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SYED VAHEEDUDDIN

1907—1960

Dr. Syed Vaheeduddin, Plant Pathologist, Andhra Pradesh, died on December 28, 1960, at the age of 53. He was born on 12th. June 1907. After his early education at Hyderabad he joined the Poona Agriculture College from where he got B. Ag. (Diploma) in 1928 after a brilliant career. Immediately afterwards Vaheeduddin joined the Department of Agriculture in 1928 in the H. E. H. Nizam's Service. After working for a couple of years he left for U.S.A. for post-graduate studies at the University of Minnesota, and was awarded the Ph. D. degree in 1936. It was here that he was associated with Dr. Elvin C. Stakman, an association which resulted in Dr. Vaheeduddin attaining international fame in Plant Pathology. His work on "Hybridization and Segregation in crosses between *Sphacelotheca sorghi* and *Sorosporium reilianum*" and "Pathogenicity and Genetics of some Sorghum smuts" is outstanding. He demonstrated that there are a great many bio-types within *Sphacelotheca sorghi* and that they may cross freely with each other within limitations imposed by sex groups and he actually succeeded in synthesizing a new parasitic race of *S. sorghi*. Most significant, however, is the fact that Dr. Syed Vaheeduddin succeeded in crossing *Sphacelotheca cruenta* with *Sorosporium reilianum* and as a result, he obtained intergeneric fertile hybrids which exhibited tremendous hybrid vigour, the hybrids resembling somewhat *Tolyposporium filiferum* in some respects. In addition, he recorded two more distinct races of *S. sorghi*. During his stay at Minnesota he was elected as a member of the Society of Sigma Xi. After returning to Hyderabad in 1936 he continued as Plant Pathology Assistant till 1939 when his good work and merit was recognised and was promoted as Assistant Plant Pathologist. Further recognition came his way when a full fledged Plant Pathology section was started in 1945 and Dr. Vaheeduddin became the first Plant Pathologist of the erstwhile Hyderabad State. As Assistant Plant Pathologist, he conducted a phytopathological survey of the State during 1939-1945. This survey was the first of its kind in any Indian State. When the Plant Pathology section was started, he worked hard to put the section on a firm footing and gradually spread the activities to various aspects of Plant Pathology. He evolved ST. 1, ST. 2, ST. 3, *tur* varieties, which are resistant against *Fusarium* wilt. Similarly he put Hyderabad on the map of wheat rust work in India by evolving Hyderabad wheats (HW. 1, HW. 2, HW. 3,) which are highly resistant to the physiologic races of black and brown rusts occurring in Deccan. With the formation of Andhra Pradesh in 1956, he expanded research work on plant diseases and established three Plant Pathological sub-stations for tackling the disease problems

The departmental administrative set up took away Dr Syed Vaheeduddin from active association with Plant Pathology when he was promoted as Headquarters Deputy Director for Research in November 1959. Probably this did not go well with his mental approach which was that of a true scientist. Many times he had expressed his unhappiness for being "away" from Plant Pathology. He was suffering from high blood pressure. But it was the bursting of appendix that brought his sudden death at M.G.M. Hospital, Secunderabad.

Dr. Vaheeduddin published about 40 papers covering a wide field. He was a member of Plant Diseases Committee of the Indian Council of Agricultural Research for a number of years. Also he was a member of the Indian Botanical Society as well as the Indian Phytopathological Society.

The name of Dr. Vaheeduddin will be long remembered as a gentleman and a true Scientist whose contribution to Science is much acknowledged. He was one with the motto "Work is worship" and his sudden and unexpected demise will be lamented by his innumerable friends and followers in India and abroad.

Dr. Syed Vaheeduddin leaves behind his wife and two daughters.

P. Govinda Rao.

PRESIDENTIAL ADDRESS

RECENT ADVANCES IN THE CONTROL OF PLANT DISEASES*

T. S. RAMAKRISHNAN

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Diseases exact a heavy toll of crop yields and cause considerable damage to crops year after year. Diverse methods have been adopted from time to time to mitigate the ravages caused by these diseases with varying amounts of success. The acute shortage of food supplies in several countries and the rapid increase in population have, in recent years, brought to the forefront the dire necessity to conserve as much of the crop yields as possible by reducing the losses caused by diseases.

The problem of the control of plant diseases has been engaging the attention of pathologists all over the world for many years and is being tackled from different angles. On this occasion I propose to confine myself briefly to the use of fungicides and their method of application. Following the discovery of Bordeaux mixture by Millardet in France during the last century considerable attention has been devoted to the study of the mode of action of the fungicide on the pathogen. Many new formulations classified as 'insoluble coppers' have been placed on the market as substitutes for Bordeaux mixture. Yet the latter is still in strong favour in India and many other parts of the world. In South India it is in great demand for the control of the leaf rust of coffee, abnormal leaf-fall of rubber, fruit rot of areca, etc. Over 1,000 tons of copper sulphate are utilised every year in this region alone. Throughout the world the same fungicide is being widely used for spraying potatoes, grape vines, banana, coffee and various vegetables, ornamentals and fruits. The advantage with Bordeaux mixture is that the deposit on the leaves acts as a persistent protectant not readily washed away by rains. Further, definite stimulatory effects have been noticed on some crops like coffee, potato and grapes even in the absence of disease. But its preparation is laborious and a little exacting, and large quantities of the fungicide are required for affording adequate protection. Moreover, on certain varieties of crops it has a tendency to be phytotoxic.

The new 'insoluble coppers' include compounds like tribasic copper sulphate, copper oxychloride, cuprous oxide, copper phosphate and others. Of these, however, the first three alone are used on a large scale at the present day either as dusts or as water misible spray formulations. But they lack the tenacity of Bordeaux mixture and can be successfully used only in regions of lower rainfall and where repeat applications are carried out periodically. India was importing all her requirements of copper sulphate and other copper fungicides for many years. But in the last

*Presidential Address delivered at the Thirteenth Annual General Meeting of the Indian Phytopathological Society held at Roorkee, January, 1961.

decade factories have been established in more than one region in the country where both copper sulphate and copper oxychloride are being produced.

The older home-made spreaders and adhesives like resin-soda and vegetable oils which were being added to Bordeaux mixture to improve its stickiness have now been found to be unnecessary and sometimes adversely affecting the efficacy of the mixture.

Organo-copper compounds have also been tested as fungicides. But these are not generally suitable for use in live plants. Organic copper compounds like copper naphthanate, copper oleate and copper 3-phenyl-salicylate have been employed as wood preservatives, for rot-proofing cordage and in textile industry.

Next to copper compounds, pure sulphur and sulphur compounds (lime sulphur) have been popular fungicides for a long time. These were preferred for the treatment of powdery mildews and for the protection of those crops which were sensitive to Bordeaux mixture. Even sulphur has phytotoxic effects on certain crops (melons) and causes leaf-scorch especially in hot weather. But it is unique among fungicides in that it need not come into actual contact with the pathogen to exert its fungi-toxic effect. The exact manner in which it become toxic to fungi is becoming clear as a result of recent work. It is also known that the fungicidal properties of sulphur are improved by the reduction in the size of the particles of the powder. Consequently, definite specifications have been laid down for particle size (325 mesh) in fungicidal sulphur powder. Water-miscible sulphur formulations (wettable sulphur) have also been placed in the market for use, where protection is to be achieved by spraying. Sulphur powder is used for dusting crops against rusts and powdery mildews and for seed treatment of sorghum against grain smut. The insecticidal properties of sulphur particularly favour its use in particular instances as for the control of *Oidium* and mites on rubber. Sulphur is also one of the constituents of some of the new organic fungicides. One snag in the widespread use of sulphur (pure powdered) in India is in the restriction placed by the Government on its purchase, movement and storage. Processed sulphur containing a mixture of 30 to 40 per cent of talc has been found to be as good as pure sulphur in the control of certain powdery mildews like that on rubber and such processing will be helpful in getting over the difficulty mentioned above. But sulphur itself is in short supply in our country and has to be imported from abroad.

In earlier years inorganic mercury compounds (e. g. mercuric chloride) were used on a limited scale as fungicides. The cost of the material and the highly poisonous nature of the formulations precluded their use on a large scale. But after World War I, several organomercury compounds came into use especially for seed treatment. Uspulun and Germisan were among the earliest of these from Germany. These were followed by various other formulations like Ceresan, Agrosan etc. and at the present day are used all over the world as seed disinfectants and protectants. Some of these have been employed for field treatments also,

against apple scab and rice blast. Amongst other uses for some of these are for the treatment of soil-borne diseases in vegetables, sugarcane and rubber (in Ceylon). In South India two of them have been successfully used for the control of panel diseases of rubber.

The organo-mercurials used as fungicides are ethyl mercury compounds. They are used as dusts or water-miscible formulations containing a very low percentage of the active ingredient. As they are poisonous to human beings, the consumption of the treated parts as food should be done only after careful washing of the material. Besides their fungitoxic action other beneficial effects on the host plant have been recorded for some of them though this has not been conclusively established in all cases. When used for the control of the panel diseases of rubber initial increases in the output of latex have been observed. All the organomercurials used in this country are imported from abroad at the moment. Any attempt at producing them in the country will make them easily available to the farmer and may reduce the cost also.

The high fungitoxic action of silver and nickel compounds has been reported by several workers from the results of small scale experiments. They have not come into extensive usage presumably due to their non-availability and prohibitive cost.

During and after the World War II, probably urged by the need of copper and mercury for other purposes, several organic fungicides have been developed and placed on the market. Some are reported to be more efficient against specific diseases than the older fungicides. These belong to the group of carbamates, quinones, phenols etc. Some of them are used for seed treatment, like phygon and spergon, while others are employed for field applications. A few have a wide range of use, like captan and Dithane. Nabam and zineb were introduced in the United States with great advantage for the control of the late blight of potato. The advance in the knowledge of the mode of action of these chemicals on fungi has led to the formulation of new compounds possessing greater and more persistent protective effects. The modern tendency is for formulations having specific uses than for a fungicide with a wide range of uses. Greater scope for further improvement exists in the field of organic fungicides than in the older inorganic ones. Formulations of organic nemacides and soil fungicides like mylone and vapam are also coming into use.

But most of these developments have taken place in foreign countries like the United States, the United Kingdom or Germany and consequently these substances are very costly and not readily available to the farmer in India. Till their utilisation is brought within the reach of the farmer's capacity, their use in India may be confined to a few experimental trials by certain institutions and not become widespread.

The pious hope of many pathologists is for the development of systemic fungicides which will prevent infection of the host or destroy the pathogen alone inside the host as a result of its application on the folige or to the roots. The protective fungicides that are in use today are not

suited for this purpose as they do not penetrate into the tissues or become immobilised as soon as they enter them. Further, they may remain innocuous when applied externally but they may become phytotoxic once they are inside. Certain antibiotics have been found to penetrate and become systemically distributed in some plants (streptomycin). Some bacterial plant diseases have been claimed to be controlled by the use of streptomycin (fire blight of pears, bean blight and canker of citrus). However, the benefits appear to be due to the immediate antibacterial action of the antibiotic rather than to any systemic action. There is also the possibility of producing resistant races of the pathogen by the continued use of the antibiotics. Actidione and anisomycin have been used for controlling powdery mildews and bean rust, respectively. Another antibiotic which has received much attention is griseofulvin. In all these studies, sustained systemic effects have not been obtained against fungal diseases. The investigations are still in progress and it is premature to make any categoric statement about these substances as systemic fungicides. The ideal systemic fungicide has to persist sufficiently long in the tissues and must prevent the growth of the pathogen without adversely affecting the growth or the value of the crop. Since fungi also belong to the vegetable kingdom the prospects of finding a systemic fungicide harmful to the fungus inside the tissues but not to the host plant are remote.

There has been considerable progress in the improvement of the equipment for application of fungicides on crops. In the early decades of the century, field treatments consisted of high volume spraying with hand or power operated machines. The formulations were distributed through nozzles at high flow rates. This necessitated the use of large volumes of water. Very often the non-availability of water or its transport from long distances interfered with the adoption of protective measures in time. The use of dust formulations of fungicides was tried to get over this difficulty. But except in the use of sulphur, dust applications were invariably found to be less effective and, in some cases, of no value for the control of diseases of crops. Intensive research on the development of new types of nozzles and other equipment aiming at distributing low volumes of fungicides over large areas followed. We have now several types of low volume nozzles suitable for hand operated and power operated sprayers. Mist blowers have also been developed in which a blast of air helps in the atomisation and distribution of the fungicides in the form of very fine droplets. In this process a higher concentration of the active chemical could be employed though the total quantity used is comparatively less. The carrier of the fungicide is often not water but some non-phytotoxic grade of mineral oil. The water drops blown through a mist blower evaporate even before they reach the plant surface and the fungicide is not deposited satisfactorily on the plant, where as the drops of oil remain as such and adhere to the leaf well. The coverage is satisfactory and there is no run off. But the mist blowers are all of foreign manufacture and cost much so that they can be availed of only by richer farmers.

Oil-based fungicides and sometimes oil alone have been applied on a large scale in banana plantations in the West Indies and Australia for

the control of the 'shigatoka' disease. For the last three years oil-based copper oxychloride has been used with remarkable success in South India for the control of the 'abnormal leaf-fall' of rubber. The fungicide is applied from the ground with a power operated Micron 420 model sprayer. This method is bound to be favoured by large holders as the alternative Bordeaux spraying is a high volume operation requiring very large quantities of water, not easily available during the spraying season (April-May). Further, the new method obviates the necessity of climbing the trees at great risk. But again these machines can become popular only when they become readily available.

Helicopters and fixed wing aircraft are being pressed into service for the application of fungicides over large areas in quick time. Much success has not been achieved by this method in the control of fungal diseases as against pests. Fog machines, though reported to be useful against pests, have not been satisfactory for controlling diseases.

A new types of duster called the 'Electroduster' is claimed to be capable of utilising the differences in the nature of the electrical charges on the leaf surface and on the dust particles applied through the duster for making the dust stick firmly to the leaf surface. However, the trials carried out in Ceylon with this machine for dusting sulphur for controlling *Oidium* on rubber have shown that it was in no way superior to the ordinary power duster.

In recent years considerable progress has been achieved in the development of new and effective fungicides and in the improvement of the machinery for their application. The most significant advance has been in the understanding of the mode of action of several of the fungicides which is bound to result in extended and systematic studies of new and specialised formulations. The development of organic fungicides has placed a fruitful group of chemicals for further improvements in the hands of the organic chemist. The problem of disease control has to be tackled as a team-work in which chemists, pathologists, engineers and farmers have to work in close collaboration. This is much more imperative in India which is a great agricultural country and in which diseases of crops claim a high proportion of avoidable losses.

CORYNESPORELLA: A NEW GENUS OF HYPHOMYCETE

R. L. MUNJAL AND H. S. GILL

(Accepted for publication February 10, 1960)

During routine collections of Plant Disease Specimens, in the Manali and Kulu areas of Kulu Division (5,000–6,000 feet a.s.l.), Punjab an interesting Hyphomycetous fungus growing saprophytically on dead stems of *Urtica dioica* in a humid location was collected. The fungus forms distinct and conspicuous colonies on the substratum. The colonies are dark brown to black, hairy, irregularly scattered in patches, sometimes confluent and of variable size. The mycelium is partly superficial and partly immersed in the substratum, composed of branched, septate, subhyaline to olivaceous brown, smooth-walled, 3–7 μ thick hyphae. The hyphae are closely septate and are intercellular appearing knotted at places. The conidiophores arise singly or in groups of 2–3 directly from mycelium. Conidiophores are erect, straight or somewhat flexuous, dark brown, septate, thicker at the base (19–25 μ), tapering towards the tip (13–19 μ in the middle and 11–14.5 μ at the apex) and upto 1048 μ long with a slightly swollen and rounded base. These appear to the naked eye as black hair-like bristles. Upper one third part of the conidiophore is fertile and bears secondary and tertiary branches, the cells of which may again proliferate to produce conidia at their apex through the apical pore (Fig. 1a). The branches arise as lateral appendages in the upper cell of the conidiophore just above the septum through a pore in the cell wall. The branches become curved to run parallel to the main stalk. The terminal cell of the conidiophore continues to proliferate, thus producing 3–4 side-branches as also bears conidium at the apex through the apical pore. The side-branches may again produce tertiary branches giving the head a penicillate appearance. The scar (pore) at the base of the branches is very prominent and is 2–3 μ in diameter. The branches are concolorous with the stipe or slightly dilute coloured and measure 46–90 μ in length and 9–11 μ in width and easily break away from the conidiophore with the slightest pressure. The conidia (Fig. 1b) are straight or flexuous, subcylindric to obclavate, subhyaline to pale olivaceous, smooth walled, pseudoseptate, 98–270 μ long and 7–12 μ thick with a dark truncate scar at the base 7–9 μ wide.

The noteworthy features of the fungus are (i) Proliferation of the conidiophore through the apical pore with successive formation of a multiseptate conidium (ii) Proliferation of conidium through the terminal cell to produce a similar secondary conidium (iii) Production of secondary and tertiary branches on the upper part of the conidiophore giving it a penicillate appearance. The fungus is best classified in the Dematiaceae-Phragmosporae. The first two characters are shared by *Corynespora* Gussow (Wei, 1950) and the third character resembles somewhat *Dendryphiopsis* Hughes and *Brachysporiella* Batista. *Corynespora* does not possess the characteristic penicilliate type of branching of the conidiophore and is, therefore, distinct. In *Dendryphiopsis*, the conidia are never in chains

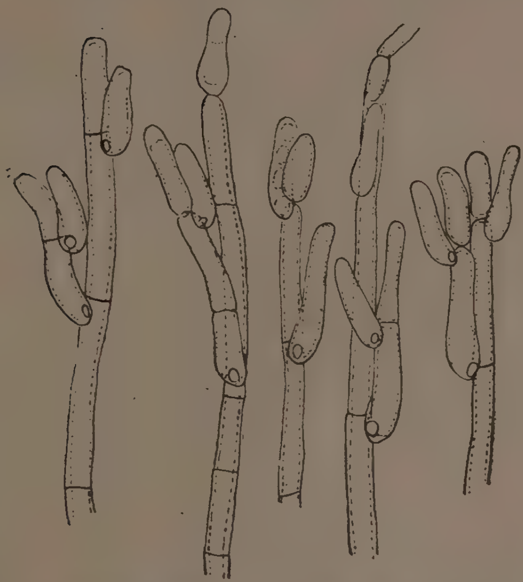

Fig. 1(a) *Corynesporella urticae*—Conidiophores showing typical branching.

and have true septa. while in *Brachysporiella*, the mode of growth of the conidiophore is totally different and the spores have also true septa. The genus *Atractina* Hoehnel (Hughes, 1953) has been shown to be synonymous with *Sterigmatobotrys* Oud. (Hughes, 1958). We know of no other genus in which this fungus can be properly fitted. We therefore propose a new genus to accommodate it and name it *Corynesporella* on account of its affinities with *Corynespora*.

Corynesporella gen. nov.

Fungus imperfectus, moniliales, dematiaceae, phragmosporae; conidiophores dark coloured, septate, erect, simple with cylindric proliferations through apical pore, secondary and tertiary branches formed on the terminal part of conidiophores; conidia formed through a pore at the apex of conidiophore as well on side branches, subhyaline to pale brown, pseudoseptate, subcylindrical to obclavate, smooth-walled sometimes proliferating to form similar secondary conidia.

Corynesporella gen. nov.

Pertinet ad Fungos Imperfectos, ad Moniliales, Dematiaceas, Phragmosporas. Conidiophori fusce colorati, septati, erecti, simplices, ornati proliferationibus cylindricis per porum apicalem; rami secundarii et tertiarii efformati in parte terminali conidiophororum; conidia efformata

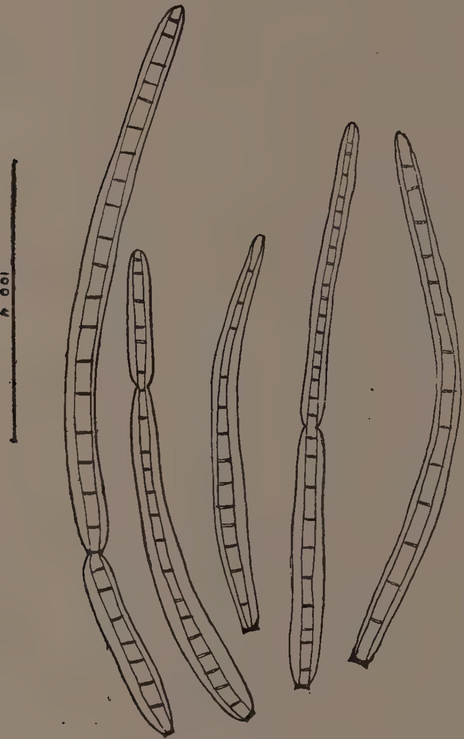

Fig. 1(b). *Corynesporella urticae*—Showing proliferation of conidia.

per porum ad apicem conidiophororum aeque ac in ramis solidis, subhyalina vel pallide brunnea, pseudoseptata, sub-cylindrica vel obclavata, levibus parietibus praedita, nonnumquam proliferantia e conidiis secundariis similibus.

Corynesporella urticae sp. nov.

Colonies dark brown to black, hairy, effused; mycelium immersed in the substratum, composed of branched, septate, pale to brown, smooth-walled hyphae; conidiophores arising singly or in groups of 2–3, erect, simple, branched terminally, straight or somewhat flexuous, pale brown to dark coloured, septate sometimes with one or two successive proliferations, 786–1048 μ long, 11–14.5 μ broad near the apex, 13–19 μ wide near the middle and 19–25 μ wide near the base; side branches usually cylindrical or oblong, 0–2 septate, 47–90 x 11 μ; conidia formed singly or in chains through a pore at the apex of conidiophore or side branches, straight or flexuous, subcylindrical or obclavate, smooth-walled, subhyaline to pale brown, 5–29 pseudoseptate, 98–270 μ long, 7.0–12.0 μ wide (7–9 μ at the scar).

On dead stems of *Urtica dioica* L., Manali, Kulu valley, Punjab, December, 1957, Coll. H. S. Gill, Herb. Crypt. Ind. Orient. No. 26828, Type.

Corynesporella urticae spec. nov.

Coloniae fusce brunneae vel nigrae, pilosae, effusae; mycelium substrato immersum, constans e hyphis ramosis, septatis, pallidis vel brunneis, levibus parietibus. Conidiophori emergentes singuli vel bini ternive, erecti, simplices, terminaliter ramosi, recti vel plus minusve flexuosi, pallide vel fusce brunnei, septati, nonnumquam una vel bina proliferatione successiva, 786–1048 μ longi, 11–15 μ lati prope apicem 13–19 μ lati ad medium, 19–25 μ lati ad basin: rami laterales ut plurimum cylindrici vel oblongi, 0–2 septati, 47–90 x 11 μ; conidia efformata singulariter vel catenulata per porum ad apicem conidiophororum vel ramorum lateralium, recta vel flexuosa, subcylindrica vel obclavata, parietibus levibus, subhyalina vel pallide brunnea, 5–29 pseudoseptata, 98–270 μ longa, 7–12 μ lata, 7–9 μ lata ad cicatrices.

Typus lectus in culmis emortuis *Urticae dioicae* L. ad Manali, in Valle Kulu, in provincia Punjab, mense decembri anni 1957 a H. S. Gill et positus in Herb. Crypt. Ind. Orient. sub. numero 26828.

We are grateful to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for his keen interest, helpful criticism and encouragement and to Dr. B. L. Chona for valuable guidance. Our thanks are also due to Dr. C. V. Subramanian for kindly glancing through the slide preparations and to Rev. Fr. Dr. H. Santapau for kindly rendering the latin translation.

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EFFICACY OF DIFFERENT FUNGICIDES. III SEED DISINFECTION IN RELATION TO DAMPING-OFF OF CHILLIES (CAPSICUM ANNUUM LINN.)

Dharam Vir and J. S. Grewal

(Accepted for publication June 10, 1960)

Chilli is an important cash crop of India and is extensively cultivated over an area of about 1.4 million acres. A large quantity of dry chilli is consumed as a spice within the country and part of it worth over 2.5 crores of rupees is exported every year. This crop is attacked by a number of diseases but 'damping-off' is a serious disease in the seedling stage and causes considerable losses to the growers. The loss can be divided into pre-and post-emergence damping off. The pre-emergence is characterized by the rotting of the seeds prior to germination or death of the germinated ones before they actually come out of the soil, while in post-emergence the seedlings are attacked by the fungus after their emergence at ground level resulting in the death of the plant.

The disease has been observed in severe form in the nursery beds of Farm area of the Indian Agricultural Research Institute during the past few years. Isolations and pathogenicity tests carried out revealed that under Delhi conditions *Pythium de baryanum* Hesse is mainly responsible for the disease. Attempts were, therefore, made to control damping-off by seed treatment with different fungicides which included a variety of organo-mercurials, quinones, dithiocarbamates, inorganic copper salts and a heterocyclic nitrogen compound.

Experimental: Steam sterilized soil was inoculated at the rate of 2.5 per cent by weight with sand maize meal cultures of *Pythium de baryanum* and the fungus was allowed to incubate in it for seven days.

The chilli seeds (variety 46A) were treated with twenty-two different fungicides. In order to ensure even distribution of the fungicides on the seed, the fungicide and the seeds were shaken in flasks on a mechanical shaker for 15 minutes. Treated seeds were then sown in pots containing inoculated soil. Each replicate was sown with 250 seeds, there being four replicates for each treatment. Pots of equal size were used and as far as possible similar conditions of irrigation were maintained and pots were kept in the pot culture house.

The germination of the seeds was recorded after emergence. The seeds which did not emerge were recorded under pre-emergence damping-off while the data for post-emergence damping-off was collected on the basis of germinated seedlings. The final stand which is based on the total number of seeds sown, indicates that the plants have survived both pre- and post-emergence damping-off. The experiment was repeated for two years. The doses of fungicides used, data regarding pre-and post-emergence damping-off and final stand of seedlings are provided in the table.

Table showing effect of seed-dressing fungicides on pre-and post-emergence damping-off of Chillies.

S. No.	Fungicide	Active Ingredient	Dose	Percentage damping-off pre-emergence 1958	Percentage damping-off pre-emergence 1959	Percentage damping-off Post-emergence 1958	Percentage damping-off Post-emergence 1959	% Final stand 1958	% Final stand 1959
1.	Copper carbonate	copper carbonate	0.3 %	46.3	45.6	71.7	39.0	15.2	32.7
2.	Cuprous oxide	cuprous oxide	0.5 %	41.3	43.7	50.1	31.4	29.3	38.6
3.	Tillex	ethylmercury chloride	0.2 %	51.0	53.9	61.6	44.2	18.8	23.2
4.	N. I. Ceresan	ethylmercury phosphate	0.05%	33.5	28.9	28.4	20.7	47.6	56.4
5.	Fusariol	ethylmercury cyanide	0.25%	38.9	29.3	54.9	30.9	27.5	48.8
6.	Ceresan M	n-(ethylmercuri)-p-toluenesulfonanilide	0.2 %	34.3	30.3	42.3	27.3	37.9	50.7
7.	Arasan	bis (dimethylthiocarbamoyl) disulphide	0.25%	36.7	36.6	59.8	42.4	25.5	36.5
8.	Thiram	bis (dimethylthiocarbamoyl) disulphide	0.3 %	39.7	37.8	53.7	35.7	27.9	40.0
9.	Fernasan	bis (dimethylthiocarbamoyl) disulphide	0.5 %	35.3	37.3	37.3	24.1	40.6	47.6
10.	Ruberon conc.	phenylmercury acetate	0.6 %	46.1	41.5	47.4	37.6	28.3	36.5
11.	Leytosol B	phenylmercuriurea	0.05%	44.2	40.4	42.1	37.9	32.3	37.0
12.	Spergon	tetrachloro-p-benzoquinone	0.5 %	42.1	45.0	65.3	34.0	20.1	36.3
13.	Semesan	2-chloro-4-(hydroxymercuri) phenol	0.4 %	43.7	38.6	37.4	25.6	35.2	45.7
14.	Phygon	2, 3-dichloro-1, 4-napthoquinone	0.3 %	25.7	25.2	7.6	10.4	68.6	67.0
15.	Tritisan	pentachloronitrobenzene	0.2 %	66.7	57.4	25.5	37.5	24.7	27.6
16.	Agrosan GN	tolylmercury acetate	0.3 %	37.5	30.2	12.0	15.0	55.0	59.3
17.	Agrosan 5W	organic mercury compound	0.05%	31.1	27.3	15.5	16.1	58.3	61.0
18.	Flit 406	n-(trichloromethylmercapto)-4-cyclo-hexene-1, 2-dicarboximide	0.2 %	6.6	12.2	6.3	7.7	87.5	81.0
19.	Puraseed	n-phenylmercuriformamide; anilinocadmium lactate	0.2 %	18.1	19.2	26.1	19.3	60.5	65.2
20.	Zerlate	zinc dimethyl dithiocarbamate	0.2 %	37.7	37.2	67.0	36.8	20.5	39.7
21.	Parzate dry	zinc ethylene bisdithiocarbamate	0.2 %	38.4	43.3	55.7	35.1	27.3	36.8
22.	Panogen	cyano (methylmercuri) guanidine	2.25cc. per kg.	40.1	42.4	63.8	37.1	21.7	36.2
23.	Control	no treatment	...	40.8	54.8	78.2	42.9	12.9	25.8

The results indicate that seed treatment with some fungicides affords a reasonable protection to chillies against pre- and post-emergence damping-off caused by *P. de baryanum*. In the present study best results were obtained with Flit 406 which, in 1958 reduced the pre-emergence damping-off from 40.8 per cent to 6.6 per cent while post-emergence damping-off was 6.2 per cent as against 78.2 per cent in the control pots. Similar results were obtained with this fungicide in 1959 where pre-and post-emergence damping-off was 12.2 per cent and 7.7 per cent respectively as against 54.8 per cent and 42.9 per cent in the untreated controls. Out of the remaining fungicides, Puraseed gave good results amongst the organic mercurials while Phygon was superior among the quinones. Dithiocarbamates and copper fungicides, however, did not appear to be promising. Jacks (1954) working on the control of damping-off of spinach caused by *Pythium ultimum*, also obtained good results with Flit 406 and Phygon. Grewal and Dharam Vir (1958) obtained good results for the control of *Macrophomina phaseoli* on Jute by seed treatment with Flit 406. Cruickshank and Jacks (1953) were able to reduce pre-emergence losses of linseed and linen flax by seed treatment with Phygon.

Summary

Twenty two fungicides which included a variety of organo-mercurials, quinones, dithiocarbamates, inorganic copper salts and a heterocyclic nitrogen compound, were tried for the control of damping-off of chillies. Flit 406 [n-(trichloromethylmercapto)-4-cyclohexene-1, 2-dicarboximide] gave best control while Phygon and Puraseed were superior to rest of the fungicides for the control of the disease.

Acknowledgement: Our grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for suggesting the problem, keen interest and valuable guidance during the course of this work.

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DILOPHOSPORA LEAFSPOT OF WHEAT IN INDIA

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During the last week of April, 1959, a leafspot disease was observed on wheat variety NP 4 at Chogal, Handwara in Kashmir Valley. Later this was also collected on local wheat from Hamre, another locality in the same valley. By the third week of May, when these localities were visited again, the disease did not appear to be so conspicuous due to the emergence of new healthy leaves.

The disease is characterised by the yellow, elliptic to elongate, sometimes spindle shaped flecks which later turn tan coloured and develop black crusts in the centre. These spots appear on both the surfaces of leaf, being more common on the upper surface and sometimes covering as much as half the area of the leaf. They are mostly single, rarely coalescent, 4–8 x 1–1.5 mm. and delimited by the mid rib; at first covered by epidermis but later exposing the black stroma which gives a charred appearance to the spot. Badly diseased leaves dry up and may fall off. Lesions similar to those on leaves but mostly gregarious and coalescent may develop on the leaf sheaths.

Black dot like pycnidia are found studded in the stroma. They (Fig. 2a) are globose, ostiolate, dark brown with a parenchymatous wall and measure 60-120 x 55–100 μ in diameter. The ostiole is broad, measures 25–40 μ in diameter and is formed of elongated cells which are torn at the apex, facilitating discharge of the spores. Spores are (Fig. 2b) straight or slightly curved, hyaline, 0–3 septate, provided with plumed or brush like or claw shaped appendages at each end. They measure 10–17.5 x 1.5–2μ (23–33 μ with appendages).

The hyaline spores of this fungus with typical claw shaped appendages at each end are characteristic of the genus *Dilophospora* of Scolecosporae (Sphaeropsidaceae – Sphaeropsidales), which cannot be confused with any other genus. Three species of *Dilophospora* have been recorded so far on Gramineae. Description of *Dilophospora* given above compares with that of *D. alopecuri* (Fr.) Fr., which is reported to be parasitic on wheat. The other two species of *Dilophospora* have broader spores.

Dilophospora alopecuri is known to cause spiral twisting of leaf and pedicel below the ear in grasses and cereals. This twist phase of the disease was almost absent in the Kashmir; only in a few cases the leaves of the affected plants became twisted in spirals. This is probably due to unfavourable environmental conditions which did not permit the normal development of the twist phase of the disease.

Fig. I. (Diagrammatic)

a. Symptoms of disease on leaf.

b. Symptoms of disease on the leaf and spiral twisting of diseased leaf.

c. Black lesions on the leaf and leaf sheath.

d. Enlarged leaf portion showing the disease lesions more prominently.

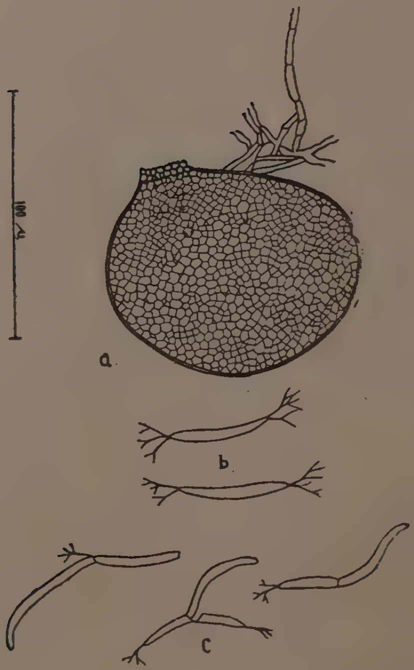

Fig. 2. *Dilophospora alopecuri* (Fr.) Fr.

a. Pycnidium

b. Spores

c. Germination of spores.

This disease is reported to occur in Europe and America. Berkeley (1862) described it on wheat in England and stated that the disease had appeared in a serious form. Subsequently also (1928–32), it was reported on wheat crop in England but not in such a serious form. Sprague (1950) maintains that the disease is sometimes of moderate importance in Europe, though it is not sufficiently important in the United States. In Germany (Anonymous, 1926), where the incidence of disease on wheat and rye was about 30 per cent, it was supposed to have been introduced on straw and seed from France and Switzerland. In a number of localities in Germany, the disease is reported to be associated with wheat nematode (*Anguina tritici*).

Preliminary observations to ascertain the source of introduction of this disease into India, were unsuccessful. It is quite likely that the disease has been present in this country for a long time but has been observed only now. The disease has not been observed on Rye or grasses so far. In view of the fact that this disease has appeared in N.P. 4, an important wheat variety grown over large areas it requires great vigilance.

The spores of the causal fungus germinate readily in sterilized water. They swell up considerably and generally break into two, each part germinating separately from the broken end or just below the appendages. The germ tube is practically isodiametric with the spore. The optimum temperature for spore germination lies between 20–25°C, though good germination also occurs at 30°C. At 15°C or 35°C however, the germination is totally inhibited. The fungus grows readily on Potato dextrose agar fortified with yeast extract and on Oat meal agar. The Pycnidia which are single and scattered are abundantly formed on the former medium after one month.

Sincere thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for his keen interest and encouragement.

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OCCURRENCE OF 'BLACKLEG' DISEASE OF CABBAGE IN INDIA

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Cabbage is an important winter vegetable in India grown over large areas in the plains. In the hills, however it is a mid summer crop, where it is sown in June and is ready by October-November.

During October, 1958 and again in October, 1959 some diseased plants were received for examination from the Vegetable Breeding Sub-station, Katrain, Kulu Valley, Punjab, located at an elevation of 6,000 feet. The disease was reported to be serious. A study of the disease on the spot showed that about 40 per cent of the plants, variety Drum-head, in certain fields were affected. The affected plants showed typical symptoms of black-leg disease caused by *Phoma lingam* (Fr.) Desm. The disease has not hitherto been recorded in India but is known to occur in Canada, U.S.A., Africa, Europe, Palestine, China, Philippines, Japan and Newzealand.

The fungus attacks roots, stem and leaves. The disease makes its appearance in the field immediately after the rains. The first apparent symptom is the wilting of young plants towards the end of August. The affected plant shows rotting of lateral roots and blackening of the tap root from tip upward. The extent of blackening depends upon the severity of attack and sometimes involves the whole root. Disease lesions are frequently observed on the stems and the leaves of adult plants, though on the young plant these are inconspicuous. Elliptic lesions are formed on the stem near the soil level which may extend in either direction and sometimes girdle it. The lesions are sunken, tan-coloured in the centre with broad purple margin and measure 5–20 x 15–30 mm. Later, numerous black dot-like, scattered pycnidia are formed on the necrotic spot. Lesions on the leaves are circular to angular in outline being delimited by veins, dirty-white with a light brown spreading margin and measure 5–20 mm. in diameter. Numerous spots may be formed on the leaves which often coalesce to form larger lesions. In humid weather, the spots are indefinite in extent and cover a large part of leaf lamina. Usually the spots are restricted to lower and older leaves and show numerous black dot-like pycnidia. In advanced cases of infection, snipping and toppling over of adult plant is a common feature.

Sections through the infected spot show that the mycelium is embedded in the host tissues. It is both inter and intracellular, at first hyaline, later olivaceous, closely septate and 4 μ in diameter. Pycnidia are not observed on the under-ground root portions. On the stem and leaf lesions, the pycnidia are subepidermal, globose or lenticular with somewhat protruding ostioles, thick-walled, wall composed of paren-

chymatous cells and measure upto 315 μ in diameter (Fig. 1a). The conidia are hyaline, single-celled, (Fig. 1b) straight or slightly curved, both ends rounded, guttulate, measuring 4–6 x 1.5–2.0 μ and are borne on tips on short cylindric hyaline conidiophores inside the cavity of pycnidia. These are exuded in a mucilaginous mass in the form of spore tendrils. Description of the fungus and the symptoms produced tally exactly with *Phoma lingam* and the pathogen is, therefore, identified as such.

The fungus grows readily on *Potato Dextrose agar over a wide range of temperature, from 15–30 °C. with optimum at 20–25 °C. The mycelium is creeping, silky-white at start, turning olivaceous later and then black after the pycnidial formation. The pycnidia are semi embedded in the substrate, black, ostiolate and somewhat irregular in shape. The spores come out as a pinkish ooze. The optimum temperature for sporulation is 20 °C and pycnidia are formed in abundance on potato-dextrose agar. These results are, therefore, at variance from those of Cruickshank (1953) who did not find the medium suitable for the growth of this fungus and reported the optimum temperature for sporulation as 25 °C. Buddin (1934) and Dennis (1939) have, however, observed the occurrence of different strains of the fungus which may explain the differences reported herein.

This is a dreaded disease of cabbage and attacks a number of related crucifers such as cauliflower, knolkhol, turnips, swedes etc. The life history of the disease has been worked out by a number of workers in different countries. The fungus is known to perpetuate through the diseased plant debris as well as infected seed. While seed infection is common in U.S.A., it is not of much importance in Britain.

The disease appears to have been introduced into India from abroad through the infected seed. The disease had been observed at the farm during the last three years and had appeared year after year in the same plots. No infection of the flowers or pods was observed at Katrain and the chances of seed infection as observed in U.S.A., therefore, were rare, possibly due to dry conditions prevailing during the flowering and the fruiting period. Seedlings raised from the same seed but transplanted into other plots did not show any infection at all. The dead, infected roots remaining in the soil are evidently the chief source of infection.

The diseased specimens and pure culture of the fungus have been deposited at Herb. Crypt. Ind. Orient. (HCIO No. 26830) and Indian Type Culture Collection (Accession No. G.C. 1122) of Mycology and Plant Pathology Division, Indian Agricultural Research Institute, New Delhi.

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*Peeled potatoes 200 gms. dextrose 20 gms. and agar 20 gms. in 1000 c.c water.

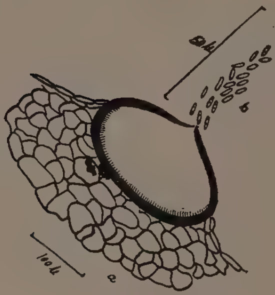

Fig. 1. *Phoma lingam*—showing pycnidium and pycnospores

Fig. 2. (a) Leaf of cabbage showing the infection spot

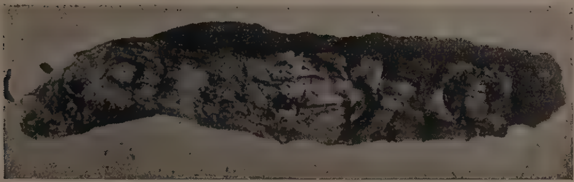

Fig. 2 (b) Diseased lesion on the stem of cabbage.

We are grateful to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his keen interest, helpful criticism and encouragement and to Dr. B. L. Chona for his valuable guidance.

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EFFECT OF VITAMINS AND HORMONES ON GROWTH AND SPORULATION OF COLLETOTRICHUM CAPSICI (SYD). BUTLER AND BISBY

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Besides the essential elements, vitamins and hormones also affect the growth and sporulation of fungi. The discovery by Wildiers (1901) of the need of a growth substance for the cultivation of yeast on synthetic media marked the beginning of studies on accessory growth substances for fungi. Marczynski (1943) observed that thiamine increased the production of dry matter by *Collybia velutipes*. In a recent work Mathur *et al* (1949) reported that certain strains of *Colletotrichum lindemuthianum* were partially deficient for one or more vitamins.

The role of hormones on the growth and sporulation of certain fungi has also been investigated by several workers. Boysen Jensen (1931) and Nielsen (1931) reported the presence of indole-acetic acid in cultures of certain fungi and bacteria. Raper (1939, 1939a) found that sexual reactions in *Achlya bisexualis* and *A. ambisexualis* were controlled by hormones.

MATERIAL AND METHODS. The investigations were carried out with single spore isolates of *Colletotrichum capsici* (Syd.) Butler and Bisby from diseased chilli plants. Czapek's medium was taken as the basal medium. In case of vitamin trials 10 μg. of vitamins were added to 100 cc. of Czapek's solution separately and in combination also. For hormone trials 0.25 mgms. of each hormone were dissolved in 2.5 cc. of 95 per cent alcohol separately. This solution was added to 1000 cc. of standard Czapek's solution and shaken thoroughly. Each solution was then distributed in 25 cc. volumes into 100 cc. Erlenmeyer flasks and sterilized at 15 lbs. pressure for 25 minutes. The flasks were then inoculated with equal amounts of inoculum and incubated at 30°C. for 21 days, and the dry weights of mycelial mats were recorded.

EXPERIMENTAL RESULTS. *Effect of vitamins.* Of the several vitamins tested, viz., Thiamine, Pyridoxine, Vitamin B12, Riboflavine, Calcium pentothenate, Nicotinic acid and Ascorbic acid, the mycelial growth of the fungus was stimulated in the presence of Thiamine, Pyridoxine and Vitamin B12. Rest of the vitamins had a depressing effect on growth. The average dry weights of the mycelial mats in milligrams were as follows:

Thiamine	237.3 mgms.
Pyridoxine	221.0 mgms.
Vitamin B12.	211.3 mgms.
Riboflavine	185.0 mgms.

Calcium pentothenate	169.6 mgms.
Nicotinic acid	164.6 mgms.
Ascorbic acid	150.3 mgms.
Control	202.0 mgms.

Sporulation was very abundant in Riboflavine, abundant in Thiamine, Vitamin B12 and Ascorbic acid and moderate in others.

The effects of different combinations of vitamins on growth and sporulation of the fungus were also investigated. The following combinations were tried: Pyridoxine+Vitamin B12, Riboflavine+Pyridoxine, Thiamine+VitaminB12, Thiamine+Riboflavine+Pyridoxine+VitaminB12, Thiamine+Pyridoxine and Thiamine+Riboflavine.

Vitamins in combination had an appreciable effect on mycelial growth. The combination of Pyridoxine and Vitamin B12 proved to be the best combination. This was followed in order by Riboflavine+Pyridoxine, Thiamine+VitaminB12, Thiamine+Riboflavine+Pyridoxine+VitaminB12, Thiamine+Pyridoxine, Thiamine+Riboflavine and Riboflavine+Vitamin B12. The dry weights of mycelial mats in different treatments were as follows:

Pyridoxine + Vitamin B12	239.6 mgms.
Riboflavine + Pyridoxine	230.6 mgms.
Thiamine + Vitamin B12	225.0 mgms.
Thiamine + Riboflavine+ Pyridoxine +Vitamin B 12	217.3 mgms.
Thiamine + Pyridoxine	215.3 mgms.
Thiamine + Riboflavine	208.3 mgms.
Riboflavine + Vitamin B 12	205.0 mgms.
Control	202.0 mgms.

Treatment differences were statistically highly significant.

Sporulation was very abundant in Thiamine + Riboflavine and abundant in Riboflavine + Pyridoxine, Riboflavine + Vitamin B 12 and Pyridoxine + Vitamin B 12. In others it was moderate.

Effect of hormones. Four hormones, viz., B-indolyl butyric acid, a-naphthalene acetic acid, Planotox, Fernoxone (Sodium salt of 2, 4-Di chloro phenoxy acetic acid) were tried for their effects on growth and sporulation of the fungus. All the hormones increased the mycelial output appreciably; statistically the treatment differences were highly significant. Of the four hormones, however, B-indolyl butyric acid gave the maximum mycelial output as compared to others. This was followed in order by a-naphthalene acetic acid, Planotox and Fernoxone. The average dry weights of the mycelial mats under different treatments were as follows:

B-indolyl butyric acid	239.2 mgms.
a-naphthalene acetic acid	230.2 mgms.
Planotox	226.0 mgms.
Fernoxone	223.2 mgms.
Control	210.5 mgms.

Sporulation was abundant in all treatments, except the control, where it was moderate.

Summary

Of a number of vitamins tested thiamine, pyridoxine and vitamin B 12 significantly increased the mycelial output. Rest of the vitamins had an adverse effect on growth. Sporulation was very abundant in riboflavine and abundant in thiamine, vitamin B 12 and ascorbic acid. In others it was moderate. Vitamins in combination had a significant effect on growth, being maximum on pyridoxine and vitamin B 12.

Several hormones were also tested for growth and sporulation. All the hormones increased the mycelial output appreciably. Sporulation was also abundant in all treatments, except the control, where it was comparatively moderate.

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RACE 122 OF PUCCINIA GRAMINIS TRITICI (PERS.) ERIKSS. & HENN.—A POTENTIAL SOURCE OF DANGER TO WHEAT CROP

R. PRASADA AND L. M. JOSHI

(Accepted for publication May 10, 1960)

Occurrence of race 122 of *Puccinia graminis tritici* (Pers.) Erikss. and Henn. in India was first reported by Gokhale and Patil (1952). Since then it has been isolated only thrice; twice from 1955–56 crop from Delhi and Kulu Valley (situated in the Himalayan Range) and recently from State of Bombay from where it was originally isolated. Obviously the race has a very restricted distribution at present, and it is not doing any appreciable damage to the crop. Its limited distribution, however, does not in anyway minimise the danger to which crops are exposed because the whole of Himalayan Range (4,000–8,000 ft. a.s.l.) is a very suitable area for the rusts to oversummer or overwinter in uredostage either on self sown plants or ratoon tillers.

The race population of a country changes from time to time and with this change, races apparently harmless and with restricted distribution assume a destructive role. Such shifts in population, involving marked variation in frequency of races have been recorded from many countries like Australia, Canada, India, Mexico, U.S.A. etc. The exact nature and the factors responsible for such changes are not fully understood but perhaps they are brought about by a number of ecological factors, micro-climate, host range etc. There are, however, a few cases on record where due to the change in varietal position certain races have assumed serious proportions. In Mexico, Gibler *et al* (1953) have attributed the sudden rise in the frequency of race 139 to the introduction of certain wheat varieties.

In U.S.A., though the presence of race 15-B had been recorded as early as 1939 and its destructiveness was well realised, no wheat variety was available in the country when it assumed a serious epidemic form. In 1950–51 it swept over North America and again took heavy toll particularly on durums in 1953 and 1954. It is, therefore, reasonable to assume that like race 15-B in U.S.A., race 122 may, at a future date, become a problem race and we have to be, therefore, prepared to meet such an eventuality (Vasudeva and Joshi, 1958).

In India most of the improved varieties, which were bred earlier against the then existing races, do not possess any resistance to this race; hence introduction of any one of them will not in anyway minimise the present impending threat from race 122. Data relating to a few important varieties, which have succumbed to the race under glasshouse tests, are set out in the following table.

TABLE. Reactions of some improved strains and varieties of wheat to individual races of Black Rust (*Puccinia graminis tritici*)

Name of the variety or selection	Races against which the variety was found resistant (0–2)	Races to which the variety was found susceptible. (3–4)	Races to which the var. showed mixed type of reactions. (0–4)
H.P. (55) – 35	15, 21, 24, 34, 40, 42, 75, 117, 194, 21-A and 42-B	122	15–C
H.P. (55) – 34	15, 21, 24, 34, 40, 75, 117, 194 and 21-A	42, 122 and 42-B.	
H.P. (55) – 13	15, 21, 24, 34, 40, 42, 75, 117, 194, 15-C, 21-A and 42-B.	122	
H.P. (55) – 11	15, 21, 34, 40, 42, 75, 117, 194, 15-C, 21-A, 42-B.	122	
NP 790	15, 21, 24, 34, 40, 42, 75, 117, 194, 15-C, 21-A and 42-B.	122	
NP 809	15, 21, 24, 34, 40, 75, 117, 194, 15-C, 21-A.	42, 122 and 42-B.	
E. 581**	15, 21, 24, 34, 40, 42, 75, 117, 194, 21-A, 42-B.	15-C and 122	
E. 2845**	15, 21, 24, 34, 40, 42, 75, 117, 194, 15-C, 21-A and 42-B.	122	
E. 3072**	15, 21, 24, 34, 40, 42, 75, 117, 194, 15-C, 21-A, 42-B.	122	
P.D. – 13 (NP 200)	15, 21, 24, 34, 40, 42, 75, 117, 194, 15-C, and 21-A.	42-B and 122	
P.D. 13 – 1	15, 21, 34, 42, 75, 117, 194, 15-C and 21-A.	42-B and 122	
P.D. – 14	15, 21, 24, 34, 40, 42, 75, 117, 194, 15–C, and 21–A.	122	

**E. 581 = 184. P. 2.–A.I.F.

**E. 2845 = (Aguilera Kenya 324) (Maroqui-Supremo) Kentana 2606 - 5Y 3C-1Y-3C-Mexico.

**E. 3072 = Egypt No. 101. Timstein 704, Ly-5y, 3i-2C (W. 178).

To meet the requirement, race 122 is being used in all glasshouse tests since 1954 and so far hundreds of indigenous as well as exotic varieties, crosses and strains have been tested against it. These tests revealed that some varieties such as Iumillo – R. L. 7(E. 930), Gaza (E. 931), Yaqui 53(Y x E–T) 2257–15–C. IC–5C–I.C. – Mexico (E. 2842), Kenya 318. A. J. 4.1 (W. 237)(E. 3132);Kenya Ploughman (E. 3133), Regent-975-11 x Gaza - 139 (E. 3158) and *T. vulgare* var. *delfii* (W. 379)–(E. 3236) are resistant under glasshouse conditions. These varieties can be used as sources

of resistance to this race and attempts have already been made to incorporate their resistance in Indian wheats by hybridization.

Summary

Race 122 of black rust, reported in 1952, is an extremely virulent race and infects practically all our improved varieties of wheat. A large number of exotic and indigenous varieties were tested with this race under glasshouse conditions and a few of them were found to be resistant to it.

ACKNOWLEDGEMENT: Our grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, for his helpful suggstions and for going through the manuscript.

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A COLLETOTRICHUM SP. ON CITRUS

K. S. Thind and G. S. Rawla

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An anthracnose or die-back of *Citrus limon* Burm. (Rough lemon = Jatti Khati) and *C. aurantifolia* Swingle (Sour lime) was observed during the monsoon months of August and September 1956–57 in the Khalsa College nurseries. This paper deals with the morphology and pathogenicity of the causal fungus. As it will appear from the following account it is different from *Colletotrichum gloeosporoides* Penz. which causes the common citrus anthracnose and probably seems to be a new species of *Colletotrichum* with falcate conidia.

When early symptoms appear, leaves fall off from the top of the branches and the main axis. The leafless twigs and stem parts die from top downwards, lose green colour, become woody and develop a white ashen membrane on their upper parts. On these dead white membranous parts are developed later on, black acervuli with long and abundant setae. (Plate 1.) The affected twigs remain attached in an upright position among the healthy green parts. This disease infects young plants mainly and is more prevalent in the nurseries during humid weather. No symptoms have been observed on leaves, flowers and fruits.

The disease appears in the month of August and can be seen uptil November, thereafter its percentage of infection declines and ultimately the affected twigs are damaged by saprophytes.

Morphology of the fungus. *Acervuli* 42–247 μ in diameter, erumpent, setose, dark black in colour, scattered or gregarious.

Setae 26–288 x 2–8 μ, both marginal and central, dark brown when young, black when old, long and pointed, broad at base, tapering upwards and becoming bristle like, septate, septa mostly 1–3, rarely upto 7.

Conidia 13–21 x 1.5 x 4.5 μ, hyaline, sickle shaped, borne singly on conidiophores, non septate, with tapering ends and without a vacuole in the centre (Fig. 1.).

Conidiophores 7.5–21 x 1.6–3 μ, hyaline, simple, cylindric, almost of the same size as the conidia, nonseptate.

The conidia start germination after two hours in a drop of tap water at room temperature when placed in a moist chamber and after four hours on an agar plate. During germination the conidium generally

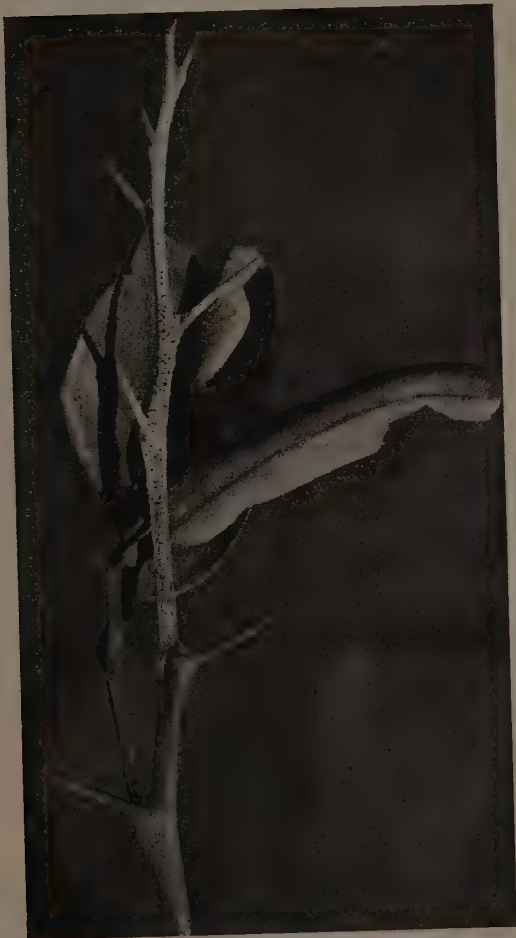

Plate I. A twig of *C. limon* Showing die-back symptoms caused by *Colletotrichum* (? sp. nov.).

becomes uniseptate, but rarely a germ tube is formed without any septation of the conidium. The germ tubes mostly appear from one or both ends of the spore or from the convex surface near the one end of the spore. The germ tubes usually swell at their ends to form a thick walled irregularly expanded structure called the appressorium (Fig. 2).

Large scale single spore isolations were obtained to have a general view of the cultural variability of the fungus. All the isolates showed the same general appearance and no variability was observed. The fungus was grown on potato dextrose agar and incubated at 28°C for six days. The colony was rounded with slightly wavy margin and showed an average diameter of 58 mm. The growth was dense and creeping, brown to black olivaceous, neither fast nor slow growing. Hyphae 1.8–7μ wide, hyaline branched, septate and thick walled. Acervuli 180–600 μ in diameter, abundant, black to buff pink coloured. Setae 76–520 x 3–8μ, abundant, dark brown to black, long and pointed. Conidia 16–35.5 x 2–4 μ, falcate, hyaline, nonseptate. Conidiophores 6.4–14.4 x 1.6–4.8μ, hyaline, cylindric. The acervuli, setae, conidia and conidiophores formed in culture are much bigger in size than those produced on the host.

An interesting feature was that some lighter coloured setae were observed to bear at the apex a single, slightly curved conidium when fungus was grown on P.D.A., thus showing that setae and conidiophores are homologous in origin and perform similar functions too (Fig. 3).

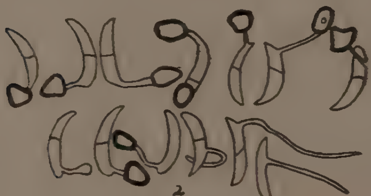

Fig. 1. **Conidia, x 1150.**

Fig. 2. **Germinating Conidia, x 500.**

Fig. 3. **Setae bearing Conidia terminally x 2530.**

The disease was reproduced successfully from the spores produced on P.D.A.

Morphologically this *Colletotrichum* sp. causing anthracnose of *C. limon* and *C. aurantifolia* is markedly different from *C. gloeosporoides* Penz. which causes the common anthracnose of citrus in having falcate conidia. In this respect it is allied to *C. capsici* which is reported to infect citrus by artificial inoculation (Butler and Bisby 1931).* Its conidia are however smaller than that of *C. capsici* while its setae are longer. Attempts to infect *Capsicum annuum* with this isolate of *Colletotrichum* species have failed so far, although *C. capsici* was able to infect the citrus hosts under similar conditions. The study carried out so far appears to indicate that this fungus is a new species of *Colletotrichum*.

SUMMARY

A *Colletotrichum* sp. was obtained during the last year in the months of monsoon causing a die-back to *C. limon* and *C. aurantifolia*. Numerous single spore isolations showed the fungus nonvariable. From the morphological characters alone, this fungus appears to be a new species of *Colletotrichum*.

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STUDIES ON THE FUNGAL FLORA OF SOME VEGETABLE SEEDS

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Several fungi found on the market seeds of vegetables are known to cause considerable damage either directly to the seeds that carry them or to the crops that are raised from contaminated seed stocks. The nature of damage consists of lowering or total failure of germination or poor development of crops showing various kinds of disease symptoms. Experience gained in Europe and America shows that at least the more important seed-borne diseases of vegetables like the anthracnose of French bean (*Colletotrichum lindemuthianum* (Sacc. & Magn.) Briosi and Cavara): blight of peas (*Ascochyta pisi* Lib.); late blight of Celery (*Septoria apii* (Briosi & Cavara) Chester) serve as a limiting factor in vegetable cultivation. Considerable progress was made in European countries with regard to the study of the seed-borne diseases of vegetable crops but in India little attention appears to have been paid to the diseases of vegetables, particularly to the micro-organisms carried with the seeds. More recently attention is being directed to the improvement of vegetable varieties in the country with a view to increase production and build up seed industry. One of the accepted methods for vegetable production is to use improved seeds. This is achieved by building up of stocks of superior quality of seeds which are true to type and free from diseases and pests. For laying down health standards against seed-borne diseases of vegetables, considerable background information is needed with regard to the micro-flora associated with such seeds, their role if any, on the disease outbreaks and the nature and extent of damage they cause. With this object in view, these studies were undertaken and data in respect of some of the commonly cultivated vegetables are reported herein.

Material and Methods: Seeds of the following vegetables namely - Cabbage (*Brassica oleracea* var. *capitata* L.); radish (*Raphanus sativus* L.); *tinda* (*Citrullus vulgaris* var. *fistulosus* Stocks); French bean (*Phaseolus vulgaris* L.); *Guar* (*Cyamopsis tetragonoloba* (L.) Taub.); Pea (*Pisum sativum* L.); Onion (*Allium cepa* L.); Brinjal (*Solanum melongena* L.); Chilli (*Capsicum frutescens* L.); *Bhindi* (*Abelmoschus esculentus* L.) which were taken up in this study were obtained from the Botany Division of the Indian Agricultural Research Institute, as well as from the markets in Delhi and from certain other seed suppliers. In some cases seeds were also obtained from actual diseased material collected from the growing crop.

For isolating the fungi, seeds showing blemishes, scars and other visible symptoms of infection were selected by examining the sample under a binocular microscope (magnification : 20x) and were germinated on moist filter paper, malt agar (2 per cent) or potato dextrose agar. When no clearly visible symptoms were observed, seeds were picked up at random. One hundred seeds were used in each of the tests carried out.

For studying externally carried seed fungi, seed washings were centrifuged and the sediment was examined. For this purpose 0.5 to 5.0 gms. of seeds, depending on their size, were added to a test tube containing 10 cc. of distilled water and plugged with cotton. After allowing the seeds to stand for 5 minutes, they were vigorously shaken and a portion of the supernatant liquid was centrifuged at 1000 r.p.m. for 30 minutes. The sediment obtained was examined under a microscope.

To examine the endophytically carried seed fungi, seeds were first surface sterilized with 0.1 per cent mercuric chloride for 2–3 minutes, washed in three changes of sterile water and sown on filter papers kept on moist wads of cotton wool or on a thin layer of malt agar or potato dextrose agar. In each Petri plate, 5 or 10 seeds, depending on the size of the seeds to be tested, were arranged aseptically at equal distances. The Petri plates were spread in a single layer on the laboratory bench. The tests were conducted during the months of November to February when the room temperature varied between 20–24°C. Petri plates were examined periodically for 12 days and as soon as the colonies came up they were transferred to P.D.A. slants. Single spore isolations were made from these cultures and all the isolates obtained were stored in a frigidaire at 10 C. for further study. Pathological tests were carried out with some of the isolates appearing to be virulent. For this purpose, 50 surface-sterilized healthy seeds were rolled on actively sporulating culture of an isolate and were placed in a moist chamber consisting of a pair of sterilized Petri dishes lined with moist filter paper. Equal number of uninoculated seeds were also kept for germination in a moist chamber side by side to serve as checks.

In a few cases, uninoculated seeds were sown in unsterilized and sterilized field soil to study the behaviour of the pathogen under natural conditions. For this purpose only those isolates showing clear indications of pathological activity during the earlier experiments were used. In each test one hundred seeds were sown. Uninoculated seeds were also sown at the same time to serve as controls.

Experimental results and Discussion: Fungi carried superficially on seeds of vegetables under study have been described. Species of *Alternaria*, *Aspergillus* and *Fusarium* were fairly common. Species of *Helminthosporium* were observed on the surface of certain seeds like radish, chilli and *bhindi*.

Endophytic fungal flora of the seeds of the vegetables listed earlier were studied on three media i.e. malt agar, P.D.A. and moist filter paper. It was observed that malt agar was the best medium for isolation of the fungal flora associated with the seed whether borne superficially or internally. Some additional fungi in certain cases were, however, isolated on P.D.A. and moist filter paper. In all 48 isolates consisting of thirteen genera viz., *Alternaria*, *Aspergillus*, *Colletotrichum*, *Chaetomium*, *Curvularia*, *Fusarium*, *Gloeosporium*, *Helminthosporium*, *Penicillium*, *Phomopsis*, *Rhizopus*, *Rhizoctonia* and *Stemphylium* were obtained and many of them were brought into pure culture. The data relating to the internally borne fungi isolated on malt agar are set out in Table I.

TABLE I. Results showing germination percentage of and fungi isolated from surface-sterilized vegetable seeds.

Seed Sample	Whether germinated or not	*Alternaria*	*Aspergillus*	*Colletotrichum*	*Chaetomium*	*Curvularia*	*Fusarium*	*Gloeosporium*	*Helminthosporium*	*Penicillium*	*Phomopsis*	*Rhizopus*	*Rhizoctonia*	*Stemphylium*	Infected seeds / Un-infected seeds	Infected seedlings / Healthy seedling
Cabbage	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/76
	Ungerminated	9	—	—	—	—	—	—	—	3	—	—	—	—	12/24	—
Radish	Germinated	6	2	—	2	—	—	—	—	—	—	—	—	—	—	10/63
	Ungerminated	4	—	—	10	—	—	—	2	—	—	—	—	—	16/37	—
Tinda	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/78
	Ungerminated	—	—	—	—	—	—	—	—	—	—	—	—	—	0/22	—
French bean	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/73
	Ungerminated	6	4	—	2	—	3	—	—	—	—	—	—	—	15/27	—
Guar	Germinated	—	—	—	1	—	—	2	—	—	—	—	—	—	—	3/92
	Ungerminated	—	—	—	2	—	—	1	—	—	—	—	—	—	3/8	—
Pea	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/86
	Ungerminated	2	2	—	—	—	—	—	—	—	—	6	—	—	10/14	—
Onion	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/69
	Ungerminated	—	—	—	—	—	—	—	—	—	—	11	—	—	11/31	—
Brinjal	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/58
	Ungerminated	3	4	—	—	—	—	—	—	3	4	1	—	2	17/42	—
Chilli	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/72
	Ungerminated	19	—	1	—	1	2	—	—	2	—	—	—	—	25/28	—
Bhindi	Germinated	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0/55
	Ungerminated	2	13	—	—	—	1	—	—	6	—	1	7	—	30/45	—

TABLE II. Results of seed inoculations carried out with fungi isolated from different seed samples. (Figures indicate percentage of infection)

Crop	*Alternaria*		*Aspergillus*		*Colletotrichum*		*Fusarium*		*Gloeosporium*		*Phomopsis*		*Rhizoctonia*		*Rhizopus*		*Chaetomium*		*Helminthosporium*	
	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings	Infected seeds	Infected seedlings
Cabbage	100																			
Radish		30	92	8													42		50	40
Tinda				50												60				
French bean					100															
Guar										20										
Pea	32	24																4		
Onion	28		44													46				
Brinjal																				
Chilli																				
Bhindi			56	16			46	26					50	46						

Seed inoculation tests with fungi appearing to be pathogenic were carried out. Data showing the effects of such fungi on germination of seeds and on the seedlings that emerged are presented in table II. *Alternaria* and *Aspergillus* were the most important isolates responsible for failure of germination and seedling damage. In addition *Fusarium* and *Rhizoctonia* on *bhindi*, *Rhizopus* on onion and *tinda* and *Helminthosporium* on radish caused failure of germination and seedling rot or blight to a great extent.

The data collected in respect of the fungal flora of different vegetable seeds are discussed below:

Cabbage: *Alternaria* was the most important fungus isolated from surface-sterilized cabbage seeds. It is one of the commonest pathogens that attacks several cruciferous crops. A number of species of *Alternaria* are reported on them. Out of these, two are concerned with black leaf spot. They are: *A. brassicae* (Berk.) Sacc. and *A. brassicola* (Schw.) Wiltshire. These fungi are world-wide in distribution and are known to be seed-borne; often they lower germination and cause shrivelling of seeds. Neergaard (1945) described *A. tenuis* Auct. a common saprophyte on germinating seeds of cruciferae. This species becomes pathogenic only slightly when the host plant becomes weak. The present isolate resembles *A. brassicae.* Infection was deep-seated and therefore surface disinfection would not eliminate it. The fungus was found in the seed coat and often it sporulated on seed surface. It brought about lowering of germination to a considerable extent.

Radish: An isolate of *Alternaria* was obtained from radish seeds also. Groves and Skolko (1944) described *A. raphani* causing leaf spot on radish. Like *A. brassicae* and *A. brassicola* it is also seed-borne. The present isolate does not appear to resemble *A. raphani* but it seems to be nearer *A. tenuis* group. The fungus produced dark linear lesions on the seedlings as they came up (Plate I).

A species of *Chaetomium* was obtained from 10 per cent of radish seeds. Contaminated seeds produced seedlings with radicles that soon became soft and light brown. The isolate brought about reduction in germination upto 58 per cent. A few seedlings that came up also showed symptoms of *Chaetomium* attack. Generally the genus *Chaetomium* is regarded as a saprophyte and is often associated with deterioration of fabrics, leather goods and paper products but Skolko and Groves (1948) reported that it also reduces germination in the case of heavily infested seed samples. Affected seeds, according to them, gave characteristic musty odour from which the presence of the fungus could be readily recognized. The present isolate also reduced germination of radish seeds but it did not emit any odour.

A species of *Helminthosporium* was isolated from 2 per cent of radish seeds. 40 per cent of germination was suppressed by this isolate during inoculation tests. *Helminthosporium* does not appear to have been recorded on radish seeds earlier but from its performance should be considered as a pathogen of considerable importance.

Tinda: On malt agar no fungal isolate was obtained from *tinda* seeds but colonics of *Rhizopus* were isolated from 19 per cent of seeds plated on P.D.A. and from 12 per cent of those sown on moist filter paper. The affected seeds failed to germinate and were completely overgrown by the fungus. 9 per cent of the seedlings that come up on moist filter paper also showed symptoms of rotting. Inoculations showed that 60 per cent of the seeds became rotten. *Rhizopus* is known to cause soft rot of several vegetables like cucumber, squash, pumpkin, musk melon and water melon belonging to the family *Cucurbitaceae.* Under favourable conditions the fungus is capable of causing considerable damage. In cultures the growth of the fungus was so rapid, that it enveloped normally germinated seeds also but it caused little damage at first. Seed contamination occurs from attacked fruits. The fungus perhaps penetrates seeds from the attacked fruit tissues and lies below the seed coat. As the affected seeds germinated, the hyphae also came out from the micropyle along with the radicles, which were enveloped by the fungus completely. Infected seeds form a source of contamination of fresh soils.

An isolate of *Aspergillus* was obtained from 5 per cent seeds of *tinda.* 4 per cent of the seedlings that came up also showed *Aspergillus* attack while 50 per cent of the sprouts showed rotting symptoms. The genus *Aspergillus* consists of some of the commonest moulds nearly all of which are saprophytes. Wiant (1937) described an *Aspergillus* rot in U.S.A. on musk melons and considered it as only one of the minor storage decays. *Aspergillus* rot is often associated with seeds showing decreased vigour. Unlike that of *Rhizopus*, the spread of the fungus is localized in cultures. There seems to be no record of *Aspergillus* on the seeds of *tinda.* Seed contamination results from decayed fruits. Infection is deep-seated and not eliminated by surface-sterilization.

French bean: Alternaria and *Fusarium* the were important fungi obtained in this case. A species of *Colletotrichum* was also isolated from seeds plated on P.D.A.

Seed inoculations with the *Colletotrichum* isolate suppressed totally seed germination. On the surface of such seeds appeared black shining masses of spores after some time. The isolate resembles *Colletotrichum lindemuthianum.* It produced setae in abundance in cultures. The ascigerous stage, *Glomerella lindemuthianum* Shear, was not observed.

The fungus produced dark spots on pods and seeds (Plate II) but these could only be seen on the seeds produced by the white-seeded varieties. Infection on the seeds from the brown-seeded varieties was on the other hand clear only when the seeds were soaked in water for some time. Infection was deep-seated and it penetrated cotyledonary tissues as well. Gram and Weber (1952) stated that seedlings arising from the affected seeds produced linear brown lesions or spots on the seedling parts and that the diseased seedlings died ultimately. But in the present tests, all the inoculated seeds failed to germinate and this shows that the symptoms vary depending on the degree of infection.

Doyer (1938) described *Macrosporium commune* Rab. on *Phaseolus* spp. and the fungus produced, according to her, pink spots at the micropylar end but she stated that infection was not deep-seated and as such was amenable to superficial treatment. There seems to be no record of *Alternaria* in India on French bean seeds.

Fusarium is another important fungal isolate obtained from surface-sterilized French bean seeds. It completely enveloped the affected seeds and after some time small stunted sprouts emerged from them. Conidia of *Fusarium* were observed in the centrifuged seed washings. This clearly shows that the fungus is also superficially carried on the seed surface. *F. oxysporum phaseoli* Kendrick P & Snyder causing yellows disease of bean was reported by Kendrick and Snyder (1942) to be carried on the seed surface. They stated that seed contamination results during threshing and it can be controlled by appropriate chemical treatment. But the internal infection, as has been observed during the present studies cannot evidently be controlled by superficial treatment.

Guar: Gloeosporium sp. is an important fungus isolated from surface-sterilized *guar* seeds. The colonies produced on the affected seeds were originally white turning later on pink owing to the formation of large number of conidial masses. It did not produce any setae on P.D.A., Recently Desai and Prasad (1955) described a *Colletotrichum* on *guar* but they did not determine the species. The present isolate does not resemble theirs.

Pea: Alternaria sp. is an important fungus isolated from surface-sterilized pea seeds. Affected seeds showed dark patches on the seed coats. Such lesions also appeared on the cotylendons of some of the seedlings. The radicles of the affected seedlings became brown and blighted. The fungus lowered seed germination considerably (32 per cent).

Onion: Rhizopus is an important fungus that was obtained from surface-sterilized onion seeds. Contaminated seeds failed to germinate. Seed inoculation tests revealed that the fungus suppressed germination by 46 per cent. Such seeds were completely overgrown by the fungus. There seems to be no record of *Rhizopus* on onion seeds from India.

Seed washings showed a species of *Alternaria* indicating the presence of this fungus on the surface of the onion seeds. Neergaard (1945) described an *Alternaria* sp. on the seeds of onion. Artificial inoculation tests showed that this isolate reduced germination by 28 per cent.

Brinjal: Species of *Alternaria* and *Phomopsis* were isolated from seed extracted from diseased fruits of brinjal (Plate III). In the first case, the affected seeds did not germinate at all and were completely covered by the mycelial growth of the fungus. Rands (1917) reported an *Alternaria* on brinjal. Padmanabhan (1948) reported *Alternaria melongenae* on brinjal from India but his account did not state whether it is seed-borne or not.

Phomopsis is an important fungus isolated from surface-sterilized brinjal seeds. *P. vexans*, the causal organism of brinjal blight was stated to be seed-borne by Walker (1952) in U.S.A. Recently in India Pawar and Patel (1957) studied the disease but they did not state whether it is seed-borne. The perfect stage of the fungus *Diaporthe vexans* Gratz was not observed in these studies.

Chilli: Isolates of *Colletotrichum*, *Alternaria* and *Fusarium* were the more important organisms obtained from chilli seeds. (Plate IV).

The isolate of *Colletotrichum* inhibited germination in the case of 8–12 per cent seeds. *Colletotrichum* causes anthracnose and ripe rot of chilli fruits. Chupp (1925) stated that *C. nigrum*, the causal organism of chilli anthracnose affected the seeds also. As such, seed disinfection is regularly practiced in some of the foreign countries to eliminate seed-borne contamination, although it involves the danger of damaging the seed. Choudhury (1957) stated that *C. capsici* (Syd.) Butler and Bisby is not seed-borne but these tests indicated that the present isolate is carried within the seeds. In nature also several times were observed infected seeds on which the fungus produced acervuli.

Alternaria was obtained from 19 per cent of the chilli seeds. Affected seeds did not germinate. Higgins (1924) stated that chilli blight organism (*Macrosporium solani* Ell. & Mart.) was seed-borne and it can be controlled by suitable chemical treatments. The present studies indicated that the fungus is endophytic and as such may not be amenable to superficial treatments.

An isolate of *Fusarium* was obtained from 5 per cent chilli seeds. All such seeds failed to germinate. Walker (1952) stated that a *Fusarium* wilt of chilli was described from Mexico and Argentine but there seems to be no record about its seed-borne nature.

Bhindi: Rhizoctonia is an important fungus isolated from *bhindi* seeds. It brought about failure of germination as well as seedling blight. The radicles of the affected seedlings became brown and rotten and numerous sclerotia were produced on them. Seed contamination by *Rhizoctonia* was reported by Baker (1947) in the case of chilli, tomato, beans and peas. Infection of seeds takes place when the maturing ovaries come into contact with contaminated soil. Andrus (1938) stated that *R. bataticola* causing ashy stem blight of beans is seed-borne and use of seeds from disease-free areas is practiced for keeping the trouble under check. Bean seed production is confined to the mid-western regions of the United States where the trouble is absent. Baker (1947) recommended hot water treatment for infected seeds for 30 minutes at 52 C.

Fusarium is another fungus isolated from surface-seterilized *bhindi* seeds. The isolate reduced the germination of seed by 46 per cent in the artificial seed inoculation tests while 26 per cent of the seedlings that came up also showed blight symptoms. Seed transmission of *Fusarium* was reported in vegetable crops like pea (Synder, 1932) and water melon (Walker, 1951). They are known to cause rotting of seeds and seedlings.

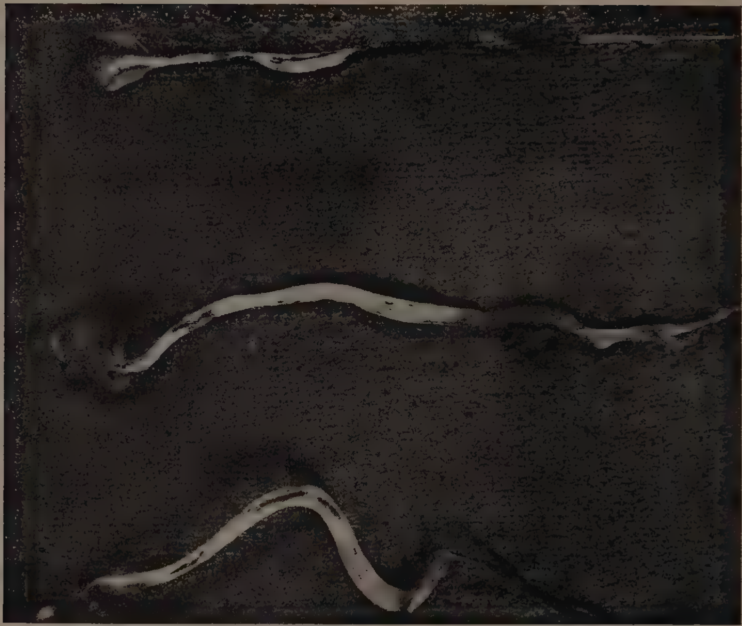

Plate I. Seedlings of radish arising from seeds contaminated with *Alternaria* sp. (isolated from radish earlier) showing blight symptoms.

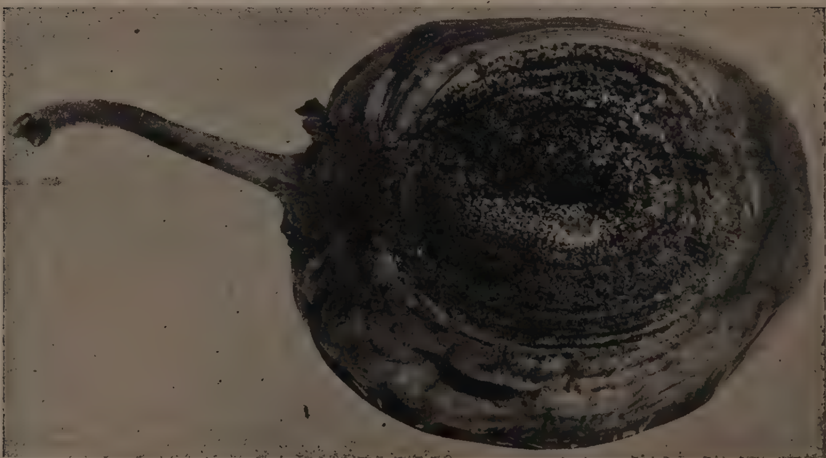

Plate III. Brinjal fruit showing a large blighted patch caused by *Phomopsis vexans*. Dark pycnidia of the fungus are seen arranged in concentric rings.

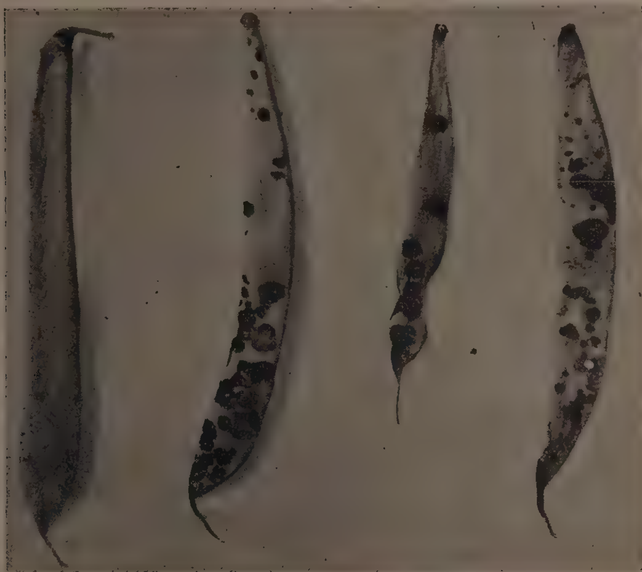

Plate II. French bean pods showing infected patches caused by *Colletotrichum lindemuthianum*. The fruit on the extreme left is healthy.

Plate IV. Chilli fruits:-

(a) Healthy
(b) Showing a large blight patch caused by *Colletotrichum* sp.
(c) Showing an *Alternaria* blight patch.

Summary

A survey of externally as well as internally carried seed fungi of ten common vegetables namely—cabbage, radish, *tinda*, French bean, *guar*, pea, onion, brinjal, chilli and *bhindi* collected from Delhi and other places was made.

Isolation of seed-borne fungi was carried out on P.D.A. malt agar (2 per cent) and moist filter paper. Malt agar was found to be the best medium for isolating many of these organisms. Of the externally carried seed fungi, *Alternaria* was found to be the commonest genus while *Aspergillus* and *Fusarium* are next in order of sequence. Important genera like *Colletotrichum*, *Helminthosporium* and *Phomopsis* are carried inside some of the seeds tested.

Artificial inoculation tests with some of the isolates appearing to be pathogenic were carried out and their effects on germination of seeds and seedling development studied.

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RELATIVE REACTION OF DIFFERENT VARIETIES OF RICE TO THE BROWN LEAF-SPOT DISEASE IN THE PUNJAB

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(Accepted for publication February 10, 1960)

The Brown Leaf-Spot disease of rice, caused by *Helminthosporium oryzae* Breda de Haan, occurs wherever rice is grown and causes considerable damage to the crop every year. In severe cases, the spots may become numerous, the entire leaves may wither and the panicles may get severely infected causing heavy sterility of blossoms. Sometimes the whole fields have been observed to present a scorched or burnt appearance. In such cases heavy reduction in yield is the inevitable result.

There are many ways to control plant diseases, but the most satisfactory and dependable one, at least for a limited number of years, seems to be the selection or the development of resistant varieties. This should be a major objective for fighting diseases, because, unless varieties that possess a good degree of resistance to them, in addition to good agro-commercial qualities, are evolved, it will not be worthwhile to recommend them to the growers.

Mere field observations to select resistant varieties may require several years, and yet the result may not be dependable. Artificial inoculation of varieties under most favourable conditions of infection is not only the most reliable method, but also the quickest.

In view of the mainly air-borne nature of the Brown Leaf-Spot disease, it was considered essential to test under conditions of artificial inoculation the reaction of a large number of rice varieties, which were available at the Rice-Breeding Station, Gurdaspur; the object being to select the resistant ones which may serve as direct introductions or may be used as parents for providing resistant genes in hybridization work.

Padwick (1954) has quoted that varieties showing different degrees of resistance have been found in Philippines by Reyes (1939), in Bengal by Ganguly (1946). in Japan by Tochinai and Sakamoto (1937), and in Columbia by Bernal Correa (1940). Ocfemia (1924) and Tullis (1935) also reported various degrees of resistance among rice varieties to this pathogen and suggested that the best means of controlling the disease was to discover or to develop resistant varieties. Cralley (1932) in America and Thomas (1941) in South India, have not, however, had sufficient success in this direction.

The seedlings of 148 rice varieties, available at the Rice-Breeding Research Station, Gurdaspur, were transplanted in a field, the distance from plant to plant being 9 inches in one foot apart rows. The conidia of *Helminthosporium oryzae* for inoculation were procured by growing the

fungus on moist sterilized paddy. For this purpose, 50 grams of seed was placed in Erlenmeyer flasks with 75 cc. of distilled water and was autoclaved for 30 minutes at 15 lbs. pressure. On the following day, the flasks were inoculated with a water suspension of conidia of the fungus and incubated at 28–30 °C. for five weeks. The inoculum was also produced on oatmeal agar and potato-dextrose agar. Sterilized water was added to the culture flasks, which were vigorously shaken and the resulting suspension of conidia and mycelia was strained through clean wire gauze into buckets containing sterilized water. Each drop of the spore suspension, when examined under the microscope, was found to contain about 30 conidia, in addition to several bits of mycelium. Artificial inoculations were made with the help of a hand sprayer in the evening during the first week of August, 1954, when it was actually drizzling.

Fig. 1. Infection rating of *Helminthosporium oryzae*, as based on rice leaves showing different degrees of spotting.

I. Immune with no spots appearing
V.R.—Very resistant with only small specks
R.—Resistant with only a few small spots
M.S.—Moderately susceptible with several small spots
S.—Susceptible with numerous large spots
V.S.—Very susceptible with numerous large spots, often coalescing.

Fig. 2. Showing the reaction of two resistant varieties, namely, China 996 and China 972 and a very susceptible variety 349 Jhona.

Three to four days after inoculation, the first symptoms of infection made their appearance. With the lapse of time, infection became more intense and showed definite spots with pale-brown centres and yellow margins on susceptible varieties.

The infection rating was based on leaves showing different degrees of spotting. The photograph of the leaves employed for evaluating the intensity of infection on leaves of different varieties may be seen in fig. 1.

I. — Immune with no spots appearing
V.R. — Very resistant with only small specks
R. — Resistant with only a few small spots
M.S. — Moderately susceptible with several small spots
S. — Susceptible with numerous large spots
V.S. — Very susceptible with numerous large spots, often coalescing

The reaction of different rice varieties to artificial infection may be seen in table 1.

TABLE 1. Showing the reaction of different rice varieties to artificial infection with *Helminthosporium oryzae* Breda de Haan

Table 1.

S. No.	Rice variety	Reaction to the disease, as based on the intensity of infection on leaves	S. No.	Rice variety	Reaction to the disease, as based on the intensity of infection on leaves
1.	Russia 894	R	46.	E.C. 2707	M.S
2.	Russia 5	R	47.	E.C. 4322	V.S
3.	Russia 9	M.S	48.	E.C. 2115	V.S
4.	Russia 889	M.S	49.	E.C. 2845	S
5.	Russia 897	S	50.	Palman 46	V.S
6.	Russia 915	S	51.	Bara 62	S
7.	Russia 2877	R	52.	Son 14	S
8.	Russia 3264	S	53.	Son 225	S
9.	China 27	V.S	54.	Magoi 378	S
10.	Rams. 20	S	55.	Mohlar 346	S
11.	No. 1031	S	56.	Mushkan 7	S
12.	China 45	M.S	57.	Kele	M.S
13.	China 47	R	58.	Charnock	V.S
14.	China 47 a	S	59.	Dular	S
15.	China 972	R	60.	Dharial	S
16.	China 988	R	61.	Marichbeti	V.S
17.	China 994	M.S	62.	Bhutmuri	V.S
18.	China 996	R	63.	Adt. 4	V.S
19.	China 1007	R	64.	Adt. 20	S
20.	China 1039	S	65.	B. 76–1	S
21.	China 1040	M.S	66.	P.T.B. 26	S
22.	China 2	M.S	67.	P.T.B. 28	S
23.	China 41	M.S	68.	P.T.B. 29	S
24.	China 42	S	69.	P.T.B. 30	S
25.	China 43	S	70.	M.T.U. 20	S
26.	China 62	M.S	71.	N. 136	V.S
27.	China 63	R	72.	Bhenibhog	S
28	No. 2 Oshu Japan	S	73.	3 M.C.	R
29.	No. 2 Senichi Japan	V.S	74.	Sutrasail	V.S
30.	Norin 8	S	75.	N. 540.	M S
31.	Norin 18	S	76.	112 M.C.	M.S
32.	Norin 6	M.S	77.	A.D.B. 4	S
33.	Norin 12	S	78.	A.D.L. 4	S
34.	Norin 36	S	79.	Co. 13	S
35.	Asahi	M.S	80.	M. 175–1	S
36.	Tai hu 65	V.S	81.	CO. 203–3	V.S
37.	Calaro 11	V.S	82.	S.M. 36–30	S
38.	Late Calaro	S	83.	A.S. 2	M.S
39.	Collusa 177	M.S	84.	W.N.D.–1	S
40.	Lady right & Calaro	S	85.	M.G.L.–2	S
41.	E.C. 2504	V.S	86.	P.T.B. 31	S
42.	E.C. 2506	S	87.	P.T.B. 32	S
43.	E.C. 2701	S	88.	H.R. 8	S
44.	E.C. 2706	V.S	89.	H.R. 19	S
45.	E.C. 2702	S	90.	H.R. 12	S

Table 1. (Contd.)

S. No.	Rice variety	Reaction to the disease, as based on the intensity of infection on leaves	S. No.	Rice variety	Reaction to the disease, as based on the intensity of infection on leaves
91.	H.R. 47	R	121.	Ramonia	V.S
92.	R. 10 Chatti	M.S	122.	Ziri Karnal	V.S
93.	R. 10 Dubraj	S	123.	Black Basmati Pathankot	V.S
94.	Type 1	M.S			
95.	Type 21	S	124.	Sethi Black	V.S
96.	Type 43	M.S	125.	Mushkan Taravri 1	V.S
97.	Type 136	S	126.	Mushkan Taravri 2	S
98.	N. 12	V.S	127.	Chahora	S
99.	N. 22	V.S	128.	Hans Raj 1	M.S
100.	Ch. 10	S	129.	Stray Plant 1	S
101.	A. 64	S	130.	Stray Plant 2	S
102.	N.P. 18	S	131.	Chalaka	S
103.	N.P. 97	S	132.	Jhona Amritsar	S
104.	N.P. 125	V.S	133.	Basmati Mixture	S
105.	N.P. 130	S	134.	Mushkan	S
106.	P.T.B. 10	S	135.	Hans Raj II	S
107.	P.T.B. 23	V.S	136.	Basmati Terkiana	M.S
108.	P.T.B. 25	V.S	137.	Mushkan Terkiana	M.S
109.	349 Jhona	V.S	138.	Basmati Dhanoa	V.S
110.	370 Basmati	M.S	139.	Jhona Dhanoa	V.S
111.	246 Palman suffaid	V.S	140.	41 Lal Nakanda	V.S
112.	278 Sathra	V.S	141.	72 Phul Pattas	S
113.	Strain 20	V.S	142.	100 Ram Jawain	S
114.	314 Jhona	V.S	143.	L. 12	M.S
115.	160 Jhona.	V.S	144.	P. 502	M.S
116.	360 Ranjha	V.S	145.	Paddy No. 17	M.S
117.	Strain 55	S	146.	Cross 1.	M.S
118.	Strain 88	S	147.	Cross 1 x Sultugar matio No. 51.	M.S
119.	Toga 28	S			
120.	I.P. 125	S	148.	Begam Local	V.S

The results of the relative reaction of rice varieties to inoculation during the summer season of 1954 (Table 1) show that most of them are susceptible to the disease. Varieties included in the resistant, or the moderately susceptible groups, are listed below:

RESISTANT: Russia 894, Russia 5, Russia 2877, China 47, China 972, China 988, China 996, China 1007, China 63, 3 M.C. and H.R. 47.

MODERATELY SUSCEPTIBLE: Russia 9, Russia 889, China 45, China 994, China 1040, China 2, China 41, China 62, Norin 6, Asahi, Collusa 177, E.C. 2707, Kele, N. 540, 112 M.C., A.S. 2, R. 10 Chatti, Type 1, L. 12,

P. 502. Type 43, 370 Basmati, Hans Raj 1. Basmati Terkiana, Mushkan Terkiana, Basmati Dhanoa, Paddy No. 17, Cross 1 x Sultugar matio No. 51.

During the summer season of 1955, the above mentioned varieties, which comprise the resistant and the moderately susceptible groups, were again subjected to severe artificial inoculation. Diseased debris infected with *Helminthosporium oryzae* was also spread over the rice seedlings before the monsoon rains started. The plot, in which these varieties were sown, was also surrounded by several rows of a few highly susceptible rice varieties. This was done to provide a constant source of highly infectious and viable inoculum for the infection of the varieties in addition to the spore suspension from bulk cultures of *Helminthosporium oryzae* used in the tests.

The results obtained by re-testing the above 40 varieties during the summer season of 1955, reveal that all the varieties tested really belong to the moderately susceptible and the susceptible groups, except the two varieties, China 972 and China 996, which are resistant. These two varieties, along with a susceptible variety, are shown in fig 2.

One of these varieties, namely China 996, has been utilized by the Economic Botanist, Cereals & Pulses, Punjab as one of the parents in hybridization work. He has crossed it with 3 Punjab varieties, viz., 349 Jhona, 370 Basmati and 246 Palman; all good yielders, but highly susceptible to the Brown Leaf Spot disease. The progenies are in the 4th generation.

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*Originals not seen.

PYCNIDIA FORMATION IN MACROPHOMA MANGIFERAE, THE CAUSAL ORGANISM OF BLIGHT DISEASE OF MANGO

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In an earlier investigation (Hingorani, Sharma and Sohi, 1960), it was shown that *Macrophoma mangiferae* Hingorani and Sharma, the causal organism of blight disease of mango, produced mature pycnidia sparsely after a prolonged incubation period (40 days) and that also on two media, namely oatmeal agar and Brown's synthetic agar medium. There was no pycnidia formation on majority of the other media tried, but, in a few cases, some immature pycnidia were observed. Incorporation of different nitrogen sources or vitamins in the basal medium, as suggested by Lilly and Barnett (1951), did not seem to aid sporulation. The use of glucose, lactose and fructose as carbon sources, however, favoured the formation of mature pycnidia to a slight degree, but not sucrose, maltose and arabinose. Even then the number of mature pycnidia formed was much less than that on oatmeal agar. There is, however, considerable evidence that exposure of fungal cultures to ultraviolet light hastens sporulation (Hutchinson and Ashton, 1930; Stevens, 1928, 1930 and 1931). It was thought desirable, therefore, to study the effect of ultraviolet radiation on sporulation in *Macrophoma mangiferae* and the results so obtained are presented in this paper.

Material and Methods. A single spore culture of *Macrophoma mangiferae* used in earlier studies was employed in this investigation.

The method of procedure, unless otherwise stated, was as follows. Full radiation from an Alpine Sun Lamp (Model IX), in which dosage could be calculated in the minimal units, was used. When the material was exposed for a period of 45 seconds at a distance of 24 inches from the arc tube, it received a minimal unit of dosage of exposure to ultraviolet irradiation, which was equivalent to 522,000 ergs per sq. cm. of the Erythema producing ultraviolet rays. The range of spectrum obtained from the lamp, as taken on an intermediate Quartz Spectrograph using an Illford special rapid plate, was found to be from 2,200Å to 6,000Å.

Experimental: Effect of period of exposure and distance of light source on pycnidia formation; The fungus was grown on oatmeal agar in Petri plates and exposed to ultraviolet rays with the covers of the plates removed. The distance from the arc of the lamp to Petri plates exposed was 40 and 60 cm. The organism was exposed to ultraviolet radiation for 10, 20, 30, 40, 50, 60 and 120 seconds at an interval of 24 hours for 7 days continuously. The first exposure was given after 24 hours of seeding. The results are given below. The sign (+) indicates pycnidia formation and (–) no pycnidia formation. The

number of (+) signs roughly indicates the relative proportion of pycnidia formed.

Period of exposure in seconds	Distance of light source in cm.	Pycnidia formation in *M. mangiferae*
10	40	–
	60	–
20	40	+
	60	–
30	40	+
	60	+
40	40	++
	60	+
50	40	+++
	60	+
60	40	++++
	60	++
120	40	++++
	60	+++

Results show that the minimum time of exposure required to stimulate the formation of pycnidia is 20 seconds at 40 cm. and 30 seconds at 60 cm. distance. In general, the number of pycnidia formed is greater at 40 cm. than at 60 cm. distance and the sporulation progressively increases with the time of exposure up to 60 seconds, after which there is no apparent difference. In the control plates, that were not exposed to ultraviolet radiation, mature pycnidia were formed after 40 days as usual, whereas in the exposed plates they were formed after 7 days.

Effect of ultraviolet light in relation to different media used on pycnidia formation: The effect of ultraviolet light on sporulation in *Macrophoma magniferae* was also studied on different media, namely oatmeal agar, Brown's agar medium, onion extract agar, carrot extract agar and mango leaf extract agar. The Petri plates were exposed for one minute every 24 hours for 7 days continuously after seeding, and incubated at 26°–28°C. The control plates did not receive any exposure.

On oatmeal agar, the aerial mycelium immediately collapsed after irradiation. The fungus completely covered the Petri plates within 5 days. As compared to the control plates, slight retardation in growth was observed in irradiated plates. Colony was regular, while the aerial mycelium was sparsed at the centre and slightly cottony at the periphery. Pycnidia were formed superficially after the fifth exposure, first around the point of inoculation and then in the other regions of growth. Pycnidiospores were also produced. Pycnidia were separate. After discontinuation of the exposure, the aerial mycelium developed again.

On Brown's agar medium, the fungus collapsed after the first exposure. The fungal growth covered the Petri plate in 6 days. The colony was regular. Aerial mycelium was suppressed around the point of inoculation and was sparse at the periphery. Ashy-grey coloured pycnidia were formed abundantly in a circular zone. Mature pycnidia were produced after 7 days of exposure.

On the remaining media, the fungus formed irregular colonies with submerged mycelium. Immature pycnidia were produced on onion extract agar and mango-leaf extract agar, but none on carrot extract agar.

EFFECT OF ULTRAVIOLET LIGHT IN RELATION TO AGE OF CULTURE ON PYCNIDIA FORMATION: Petri plates immediately after seeding (1 hour) as also 1, 2, 3, and 8 days after seeding were exposed to ultraviolet radiation for 1 minute every 24 hours for 7 days continuously. The medium used was oatmeal agar and the plates were incubated at 26°–28°C. The data are set out below:—

Age of the fungus when exposed	No. of days taken to complete growth in Petri plates (96 mm. diam.)	No. of days required for pycnidia formation
1 hour	5	6
1 day	5	7
2 days	4	8
3 days	4	9–10
8 days	Already covered	Pycnidia not formed

It is obvious that ability of the fungus to produce pycnidia quickly after exposure is directly related to its age. The older the culture, the greater the time required to form pycnidia, and comparatively an old culture did not form pycnidia at all.

EFFECT OF POSSIBLE CHEMICAL CHANGES IN MEDIA AS A RESULT OF EXPOSURE TO ULTRAVIOLET LIGHT ON PYCNIDIA FORMATION: Experiments were set up to determine whether, due to ultraviolet irradiation, some change had occurred in the chemical composition of the medium (oatmeal agar) which induced quicker and more abundant pycnidia formation. One set of Petri plates was given an exposure of 7 minutes before seeding and the other set, which served as control, did not receive any exposure. Mycelial growth was normal in both the sets and there was no pycnidia formation in either case, showing thereby the absence under these conditions of any chemical change in the medium which might be responsible for quicker formation of pycnidia.

MUTATION IN *M. Mangiferae*: Subcultures from the exposed plates were made both from the mycelial portions as well as from the regions where pycnidia formation had been induced. All these sub-cultures

were similar to the parent culture. When, however, single-spore isolations were made from the exposed cultures, a mutant strain was obtained which differed from the parent culture in having smaller spores and in its ability to form pycnidia quickly and in abundance (Plate I). The parent culture normally produced a few pycnidia on oatmeal agar after 40 days of growth, whereas the mutant strain formed them in large numbers within 7 days. The conidia of the mutant strain on the host measured 9.0–15.0 μ x 3.0–6.0 μ and in culture 14.0–21.0 μ x 3.5–7.0μ, while the corresponding measurements for the parent culture were 10.5–24.5 μ x 5.3–7.0 μ and 17.5–29.8 μ x 3.5–7.5 μ. The mutant strain has remained stable since 1954.

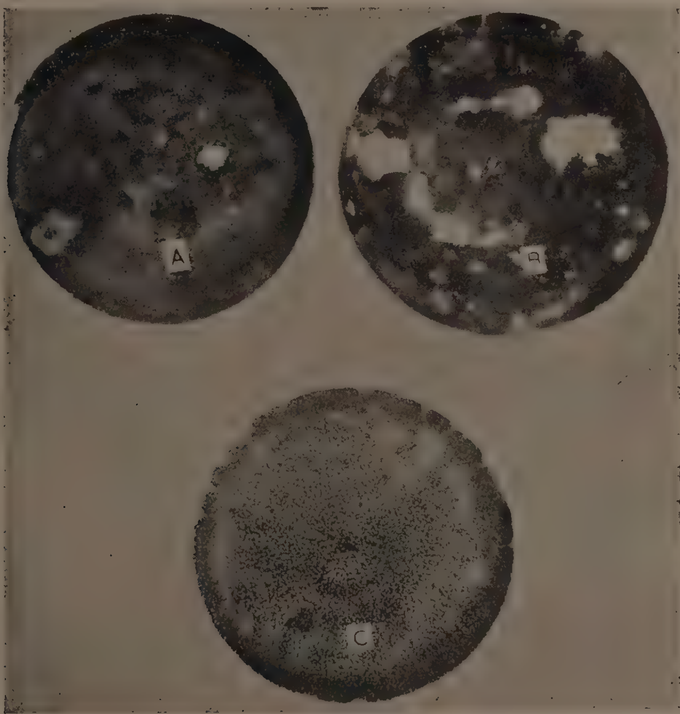

Plate I. A. Parent culture; B. Parent culture exposed to ultraviolet radiation; C. Mutant strain obtained from B by single-spore isolation.

Summary

Macrophoma mangiferae, the causal organism of blight disease of mango, normally produces mature pycnidia on oatmeal agar after 40 days of growth. It has, however, been induced to produce mature pycnidia in abundance within 7 days by exposing the culture to ultraviolet radiation.

The number of pycnidia formed progressively increases with the time of exposure to ultraviolet light upto 60 seconds, after which there is no marked difference.

Single-spore isolations from the irradiated plates have resulted in a mutant strain which is stable and is different from the parent culture in spore size and in its ability to form pycnidia more quickly and in abundance.

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ANTHRACNOSE OF COTTON IN BOMBAY STATE

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Anthracnose, one of the most destructive diseases of cotton is the primary cause of seedling blight, boll rot and fibre deterioration. Accordingly the disease is also popularly known as "seedling blight" and "boll rot" of cotton. In 1954, an epiphytotic of anthracnose was reported on *Virnar* cotton (a selection from *Gossypium arboreum* L.) from the Khandesh region of Bombay State, which grows about 7 lakh acres of this strain. Since the disease was assuming serious proportions every year and had become a limiting factor in the cotton production programme of Khandesh region, a detailed study of the disease was undertaken and the results achieved are recorded here.

HISTORICAL: Cotton anthracnose was first described from the U.S.A. by Southworth (1891) who named the pathogen *Colletotrichum gossypii* Southw. Edgerton (1909) observed the perithecial stage of the fungus on infected cotton bolls and renamed it *Glomerella gossypii* (Southw.) Edger. A seedling blight and boll rot of cotton caused by a species of *Vermicularia* was recorded from South India by Sundraraman in 1927. Duke (1927) proposed to combine the genera *Vermicularia* and *Colletotrichum* and to conserve the latter name for the genus, since it was more widely known among plant pathologists than *Vermicularia.* Dastur (1934) described an anthracnose of cotton from the former Central Provinces and Berar and provisionally named the pathogen *Colletotrichum indicum* Dast. based on the shape of conidia and its pathogenicity, which was confined to Asiatic Cottons only. In Bombay State an anthracnose caused by *Glomerella gossypii* was recorded in 1935. Uppal (1948) reported that, since the name *G. gossypii* for the fungus causing anthracnose was based on observations of the imperfect stage, it could not be definitely stated whether the fungus occuring in Bombay State was *Colletotrichum indicum* as described by Dastur.

SYMPTOMS: The disease mainfests itself twice during the growth period of the cotton crop, first at the seedling stage and again at the boll stage. On the seedlings, the symptoms usually observed are small, reddish brown spots on the cotyledons. When the atmosphere is humid, these necrotic areas encroach upon the healthy tissues until the seed leaves are entirely destroyed. At the collar, a water soaked, elongated lesion is first noticeable, which may be only on one side of the stem or may girdle it and extend downwards to the roots. As the lesion enlarges it becomes rusty brown in colour and the affected seedling collapses at the ground level and dies off. The true leaves and woody stem are not generally infected.

On the bolls, the infection occurs as small, circular, water soaked spots which later turn dark coloured and become slightly depressed. The

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spots often coalesce, forming irregular diseased areas which frequently involve the entire boll. A badly diseased boll becomes dry and mummified and finally drops off. The lint, inside badly diseased bolls, becomes discoloured and remains as a solid, compact, brittle mass of fibres. Brownish water soaked areas are noticed on the bracts which even infect the parts of the boll in contact with them. If the diseased area extends to the base of the boll, it is generally shed.

ISOLATION: The fungus was isolated from infected seedlings and bolls and was maintained on modified Czapek's agar, as it gave profuse growth and sporulation on this medium. ($MgSO_4$ – 0.5 gm. KH_2PO_4 – 1.0 gm. KCl – 0.5 gm. $NaNO_3$ – 3.0 gms. Cane Sugar – 30.0 gms. Agar – 20 gms. and Distilled water – 1,000 ml.)

INOCULATION: Healthy seeds of *Virnar* cotton were inoculated by soaking them for half an hour in a heavy conidial suspension of the fungus and were dried in air for 24 hours. These were then sown in 6″ earthen pots, filled with sterilized soil and kept in a glass house. Uninoculated healthy seeds sown in separate pots served as control. Five days after complete germination of the seeds, watersoaked lesions were observed on the cotyledons, which turned brownish in one or two days. When such a seedling was uprooted a dirty brown lesion was observed at the collar region. Infection was also obtained by spraying the seedlings and bolls "in situ" with a conidial suspension of the fungus in sterilized water. In both the cases the control plants remained healthy. Isolations made from the artificially infected seedlings and bolls yielded *Colletotrichum* sp. which resembled the original culture.

THE PATHOGEN: The fungus grew and sporulated well on Potato dextrose, Richard's Sabourad's, Oatmeal, Cornmeal and Czapek's modified agar.

MORPHOLOGY: The mycelium is slender, septate and contain droplets of oil globules. When young, the hyphae are colourless but when old they become brownish in colour. Numerous asexual fruiting bodies, the acervuli of various sizes ranging from 156 to 308 μ are produced by the fungus. The setae are olivaceous to dark brown, septate either straight or flexuous, rarely branched and pointed at the apex. The conidiophores are generally hyaline, nonseptate and measured 9.8 to 15.7 μ in length and 1.9 to 3.1 μ in breadth. The conidia are falcate, nonseptate and generally pointed at both the ends. They are hyaline when single having one or two vacuoles, but are pale pink in sporodochia like mucilagenous masses. On Czapek's agar they measure 19.1 to 30.7 μ in length and 2.8 to 4.5 μ in breadth. On the host in nature, they measure 19.7 to 32.3 μ in length and 3.2 to 4.8 μ in breadth.

TEMPERATURE GROWTH RELATION: The fungus grew and sporulated well between 26° and 31°C. No growth was obtained below 10° and above 37°C.

UTILIZATION OF CARBON COMPOUNDS: Good growth of the fungus was obtained in the presence of glucose, sucrose, levulose, raffinose and

inulin with the production of profuse mycelium; but the best sporulation was obtained in the presence of glucose, sucrose and levulose.

UTILIZATION OF NITROGENOUS COMPOUNDS: The organism is capable of utilising nitrogen from a variety of sources for its growth and sporulation of which, potassium nitrate, sodium nitrate, peptone, asparagin, creatine and DL. alanine aided good sporulation.

ENZYME PRODUCTION: The production of extracellular enzymes by the fungus in culture media was studied as per methods laid down by Crabill and Reed (1915). The results indicated that the fungus is capable of producing diastase, cystase, inulase, amydase, emulsin, gelatinase, trypsin and erepsin as exo-enzymes.

PRODUCTION OF TOXIC SUBSTANCE: Thomas (1940) reported that cotton seedlings placed in the filtrate from a 2-day-old liquid culture of *Colletotrichum indicum* wilted in 24 hours, indicating production of a toxic substance by the fungus in liquid cultures. Ling and Yang (1944) did not obtain evidence of any ill effect of the Chinese isolate of *C. indicum* on cotton seedlings placed in a filtrate from a 10-day-old culture grown on potato broth. Ramakrishnan (1947) reported that staling products, deterimental to cotton seedlings accumulated in cultures of *C. indicum* which were over three weeks old and grown on Richard's solution and that seedlings when placed in the filtrates of such cultures wilted in 12 hours.

In order to determine whether the isolate under study produced any toxic substance detrimental to cotton seedlings, cultures were grown in Czapek's modified solution. After intervals of 7, 15, 25 and 35 days of growth these were filtered through thick layers of cotton wool and the filtrates were separately filled in 500 c.c. flasks. Healthy cotton seedlings grown in sand were carefully removed, washed thoroughly under a jet of water and then placed in flasks containing the filtrates with their roots completely immersed. Seedlings placed in similar flasks containing sterilized Czapek's solution served as control. It was noticed that, none of the seedlings placed in the filtrates as well as in the control flasks wilted even after 48 hours immersion, thus indicating that the isolate does not produce any toxic substance detrimental to cotton seedlings even after 35 days growth in Czapek's solution. The experiment was repeated thrice and the same results were obtained.

GERMINATION OF CONIDIA: Highest percentage of conidial germination was obtained in Czapek's solution, *Virnar* Cotton boll decoction, *C*04 Cotton boll decoction, Cotton boll tissues in distilled water and tap water, at temperatures between 26° and 31 °C. No germination was obtained at temperatures below 10 °C and above 37 °C. Though C04 (*hirsutum*) cotton is resistant to anthracnose in India, the decoction of the bolls did not have any ill effect on spore germination of the fungus. Distilled water was found to be a poor substrate for the germination of conidia. Addition of 0.1 per cent sucrose, however increased the germination percentage.

Resistance of Conidia to Dessication: In order to study the effect of dessication on the viability of conidia of the fungus, sterile glass beads were smeared with the sporodochia like conidial masses obtained from a culture grown on corn meal agar. The smeared beads were then divided into three lots, transferred into sterilized petridishes and incubated at room temperature (23° – 25°C), 28° and 31°C. At intervals of 24 hours 3 beads from each treatment were separately planted in sterilized petridishes containing solidified Czapek's agar and incubated at room temperature. Observations on growth were taken 4 days after incubation. The experiment was repeated twice and the average of the results obtained are given in table I.

Table I. Effect of dessication on the viability of conidia of the fungus.

Beads placed in medium after.	Growth resulting from beads dessicated at		
	Room temperature	28°C	31°C
0 hours	+	+	+
24 hours	+	+	+
2 days	+	+	+
3 days	+	+	+
4 days	+	+	—
5 days	+	—	—
6 days	+	—	—
7 days	—	—	—
8 days	—	—	—

The results indicate that the conidia of the fungus can resist dessication without loss of viability for 3 days at 31°C, 4 days at 28° and 6 days at room temperature. Ling and yang (1944) reported that the conidia of Chinese isolate of *C. indicum*, smeared on glass slides, did not germinate in distilled water, after maintaining them at 28°C. for 24 hours. This could probably be because, distilled water served as a poor medium for spore germination.

Thermal Death Point of the Pathogen: The killing effect of wet heat upon the conidia and mycelium of the fungus was studied also. Test tubes of uniform thickness containing 10 c.c. of Czapek's solution were sterilized in the autoclave and inoculated with the spores of the fungus by means of a sterile inoculating loop. Particular care was taken to see that only spores were removed from the sporodochia like masses and transferred into the tubes. Likewise, separate tubes were also inoculated with the mycelium from a 73-day-old culture as well as from an 8-day-old culture of the fungus. Duplicates of such inoculated tubes were then subjected to constant temperatures ranging from 46° to 60°C. for 10 minutes in a water bath, taking care that the water in the bath was constantly stirred with a view to ensure a uniform temperature all over. One tube containing Czapek's solution was used for keeping the thermometer to record the temperature. The tubes after subjecting to the various

temperatures were immediately placed in cold water and when sufficiently cooled, incubated at 26° – 27°C. for 4 days. At the end of 4 days, observations on growth were recorded. The results are given in table II.

TABLE II. Thermal death point of conidia and mycelium of the fungus.

Temperature in °C		Growth resulting from		
		Conidia	Old mycelium	Young mycelium
46	...	+	+	+
48	...	+	+	+
50	...	+	+	+
52	...	+	+	+
54	...	+	+	+
56	...	+	—	—
58	...	—	—	—
60	...	—	—	—

The results indicate that the thermal death point of the conidia of the pathogen lies between 56° and 58°C. Muncie (1917) reported that the mycelium of *Colletotrichum lindemuthianum* from a 70–day-old culture grew even after exposing it to a temperature of 65°C. for 10 minutes and that the thermal death point of 29-day-old mycelium was between 50° and 52°C. The thermal death point of the old as well as young mycelium of the fungus under study lies between 54° and 56°C.

HOST RANGE: The host range of the organism was studied by artificially inoculating a number of plants other than cotton on which *Colletotrichum* species have been reported and also some of the common weeds growing in cotton fields. The plants were *Abutilon indicum*, *Acalypha ciliata*, *Agave* sp., *Allium cepa*, *Amaranthus* sp., *Andropogon sorghum*, *Calotropis gigantea*, *Capsicum annum*, *Cicer arietinum*, *Citrus sinensis*, *Clerodendron* sp., *Crotalaria juncea*, *Commelina* sp., *Cucurbita maxima*, *Curcuma longa*, *Datura fastuosa*, *Desmodium diffusum*, *Dolichos lablab*, *Glycine max*, *Hibiscus esculentus*, *Hibiscus rosa-sinensis*, *Lathyrus sativus*, *Linum usitatissimum*, *Lycopersicon esculentum*, *Medicago sativa*, *Nicotiana tabacum*, *Phaseolus lunatus*, *Phaseolus vulgaris*, *Piper betle*, *Ricinus communis*, *Saccharum officinarum*, *Sida* sp., *Solanum melongena*, *Solanum tuberosum*, *Zea mays*, and *Zingiber officinale*. It was noticed that besides cotton, the fungus could infect through wounds, leaves of *Zingiber officinale* and leaves (midribs) of *Saccharum officinarum*. Fruits of all plants inoculated "in situ" failed to take up infection. However, detached fruits of different plants including chilli, double bean, lima bean, brinjal and tomato kept under bell jar were infected by artificial inoculation through punctures. Dastur (1934) was unsuccessful in infecting through wounds, the fruits of chilli and tomato and leaves of sugar cane with *Colletotrichum indicum*. Ramakrishnan (1941, 1947) succeeded in producing rots by inoculation in *Capsicum* fruits and leaf spots in *Aristolochia bracteata*, *Zingiber officinale*, *Cicer arietinum* and *Curcuma longa*. Ling and Yang (1944) also

could infect fruits of pepper, egg plant, soyabean and cowpea with the Chinese isolate of *C. indicum*. It is not clear whether the inoculations made by the above investigators were on detached fruits or otherwise. Stoneman (1898) pointed out that inoculations made on detached fruits placed under bell jar could not be depended upon, since such fruits would be much less resistant than fruits under normal conditions and would be more in the nature of culture media. This was confirmed by the fact that unwounded detached bolls of Co4, a highly resistant variety of cotton got infected in 8 days after inoculation under a bell jar.

DISCUSSION AND IDENTITY OF PATHOGEN: Species differentiation in the genus *Colletotrichum* is mainly based on the size and shape of conidia and the pathogenicity of the isolate. Though the size of conidia is influenced by the substrate, it varies only within limits. Ramakrishnan (1947) found that the conidia of *C. indicum* measured 18 to 31 μ by 3.1 μ and that of *C. capsici* measured 19 to 31 μ by 3.2 μ which indicated that there was no significant difference in the spore measurements of the two species. The conidia of the isolate from cotton under study measure 19.7 to 32.3 μ by 3.2 to 4.8 μ, which agree closely with the conidial size of *C. indicum* and *C. capsici*. Thus, with regard to spore measurements all the three fungi seem to have a close relationship with one another. Moreover, all three of them produce falcate conidia.

Some workers have laid greater emphasis on the pathogenicity of the isolate in differentiating the species. Dastur (1934) provisionally erected the species *Colletotrichum indicum*, causing seedling blight of cotton based on the above criterion. He found that the isolate from cotton did not infect chillies, nor did *C. capsici*, the incitant of chilli anthracnose, infect cotton seedlings. Ramakrishnan (1947) reported that the isolates of *Colletotrichum* from *Curcuma longa*, *Cicer arietinum*, *Aristolochia bracteata*, *Gossypium* and *Capsicum* could cross-inoculate. He, therefore, suggested that all the above isolates should be brought under one species and according to the rules of botanical nomenclature, the name *C. capsici*, which is the earliest, should be retained.

Repeated trials conducted at Poona to bring about infection on chillies with the isolate from cotton, even through wounds, failed during the present investigations. As has been previously mentioned, detached fruits of chillies got infected in about 10 days after inoculation with a spore suspension of the fungus, by which time the fruits had already lost their turgidity and freshness. Thus the infection of detached fruits cannot be depended upon in differentiating the species, because such fruits would be more in the nature of culture media as conceived by Stoneman (1898).

Ramakrishnan (1947) reported that the isolate from *Capsicum* could be "educated" to become pathogenic on cotton seedlings by growing it on sterilized cotton seeds for a number of generations. Since the isolate from cotton under study failed to infect the fruits of chillies even through wounds unlike the isolate of *C. indicum* studied by Ramarkishnan (1947), further inoculation studies with the isolate from chillies, on cotton seedlings were not carried out.

Thomas (1940) and Ramakrishnan (1947) reported that staling products detrimental to cotton seedlings were produced in cultures of *C. indicum*. Ling and Yang (1944) did not obtain evidence to show the ill effect of the filtrate from a 10 day old culture of the Chinese isolate of *C. indicum* on cotton seedlings. The isolate from Bombay State also did not produce any toxic substance detrimental to cotton seedlings.

The foregoing considerations indicate that the isolate from Bombay State differs from the one studied by Ramarkishnan (1941, 1947) in its pathogenicity to *Capsicum* and the ability to produce toxic substance detrimental to cotton seedlings. It is, therefore, proposed to retain the name *Colletotrichum indicum* Dast., for the fungus causing anthracnose of cotton in Bombay State.

Fig. I. Symptoms of *Colletotrichum* seedling blight of cotton.

A. Healthy seedlings.

B. Diseased seedlings showing infection on cotyledons and at the collar.

Fig. II. Infected cotton bolls and lint.

Fig. III. Acervulus on cotton lint.

Summary

A detailed study on the morphology, physiology and host range of *Colletotrichum* sp. causing anthracnose of cotton in Bombay State has been made.

The optimum temperature for growth and sporulation of the fungus is between 26 and 28 °C. The conidia of the fungus can resist dessication for 3 days at 31 °C, 4 days at 28 °C and 6 days at room temperature (23–25 °C). The thermal death point of conidia is between 56 and 58 °C while that of mycelium is between 50 and 52.5 °C.

The fungus does not produce any toxic substance in culture, which is detrimental to cotton seedlings.

Based on its pathogenicity the fungus is identified as *Colletotrichum indicum* Dast.

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DISTRIBUTION AND PREVALENCE OF PHYSIOLOGIC RACES OF WHEAT AND BARLEY RUSTS IN INDIA DURING 1952-57.

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A comprehensive account of the occurrence, distribution and prevalence of physiologic races of wheat and barley rusts for a period of twenty years, beginning from 1932, has already been published by Vasudeva *et al* (1955). An account of the races picked up during the subsequent five years is presented in this article. During this period, with the increase in glasshouse accomodation, a much larger number of rust samples was analysed. Whereas in earlier years only 100 to 125 samples of black (*Puccinia graminis tritici* (Pers.) Erikss. and Henn.), brown (*P. triticina* Erikss. and Henn.) and yellow (*P. glumarum* (Schm.) Erikss. and Henn.) rusts were studied, it has now been possible to study, on an average, 350 to 400 samples annually. During the period under review a total of 1,790 samples of the 3 rusts was anlysed.

For the study of physiologic races, rust collections were received from different parts of the country. The collections were made either by the research workers themselves or by the authorities of the Agriculture Departments of different States. Apart from these methods of collection, Rust Nurseries have also been established at 60 representative stations throughout the country. In some of those nurseries, besides susceptible varieties of wheat, differential hosts of all the rusts were sown to serve as indicators to enable picking up of new races. One race of brown rust i.e. race 77, was first detected at the Rust Nursery at Pusa (Bihar) from a wheat variety "Mediterranean" which had been known to be resistant to all the then existing races in the country.

A careful study of the data collected during the last 25 years has confirmed that there is no regional distribution of races of these rusts in India, perhaps due to the absence of any effective natural barriers in the whole sub-continent. Over the greater part of Indo-gangetic plain, where maximum acreage is under wheat, all the three rusts are prevalent. In the rest of the wheat-growing area only black and brown rusts are found. In Peninsular India and Madhya Pradesh, black rust is predominent, with occasional appearance of brown rust; the yellow rust being practically absent, except in the hilly tracts of Nilgiri and Palny hills, because the critical low temperature, necessary for the development of this rust, is seldom obtained there during the wheat season.

The study of physiologic races of these rusts has been in progress in India since 1931 and so far 34 races and 5 biotypes, listed below, have been isolated. New races located during 1952-57 have been indicated below in block letters and their pathogenicity on differential hosts is supplied in

Table I. In Tables II-A, II-B and II-C the results of analysis of rust collections for the 5 year period have been provided.

BLACK RUST:	Races 15, **17**, 24, 34, 40, 42, **72**, 75, 117, **122**, 194. Biotype 15-C, **21-A**, **21-A-1**, 42-A and 42-B.
BROWN RUST:	Races 10, 11 **17**, 20, 26, 63, **70**, **77**, 106, 107, 108, and **162**.
YELLOW RUST:	Races 13, 19, 20, 31, A, D, E, F, G and H.

PREVALENCE OF RACES: According to Vasudeva, Lele and Joshi (1952), race 21 of black rust has shown marked fluctuations since 1932 when it was first isolated. After its first report this race was not met with again for the next seven years but since 1943 it gradually increased in its prevalence and in 1953-54 crop year it constituted 49.06 per cent of the total isolates.

Race 15 of black rust has behaved just in the opposite manner. It was one of the first four races to be reported from India (Mehta, 1933) and occupied first rank in prevalence and accounted for 41.9 per cent of isolates in 1933-34. It continued to be a widely prevalent race till 1943-44 after which there was a gradual decline and it was not even picked up in 1949-50, 1952-53 and 1954-55 crops. Race 42 is another important race because it is one of the very few races which infects Khapli wheat (*Triticum dicoccum*). Since Khapli wheat is widely cultivated in South India it is always considered to be a serious menance to this variety. This race has shown some fluctuations from time to time but has remained, throughout the period of 25 years, an important race. Biotype 42-B, however, was by far the most widely distributed.

Race 75 deserves special mention on account of the fact that it has not been isolated from the collections since 1937 although its single-spore culture is being maintained continuously in the glasshouse. Race 34, after being first isolated in 1939-40, was rarely met with during the next ten years. In fact it was not picked up at all in 1942-43, 1943-44, 1945-46, 1946-47 and 1947-48 crop years. It has, however, of late, shown a rising tendency. In brown and yellow rust races, there appears to have been no appreciable change in the position. In text figure, frequency of the most common races of the three rusts has been shown. Similar changes in the prevalence of races from time to time or shifts in population have also been reported from many parts of the world.

The reasons for such fluctuations are not fully known but the varietal position is obviously one of the important determining factors. An example of role of varieties in changing race-flora has been furnished from Mexico by Gibler, Narvaez and Enicso (1953). Before the sporadic appearance of race 15–B in 1951, Variety Supremo and others were considered to be rust-resistant but they succumbed to the new virulent biotype. The varieties were later replaced by Kentana and Kenya derivatives which were susceptible to race 139. With the introduction of these varieties, race 139, increased rapidly in Mexico but in U.S.A., from where it was first reported in 1932, no appreciable change was observed.

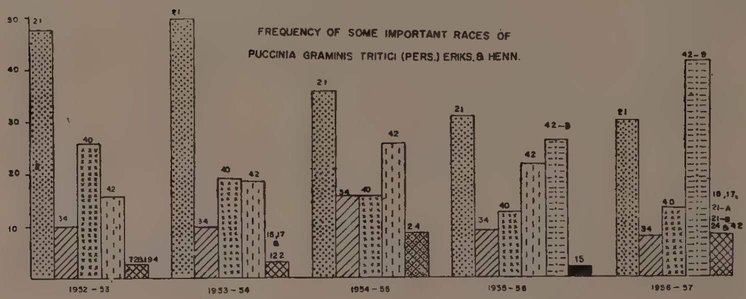
TABLE I. The reactions of differential hosts to the new races isolated during 1952-57.

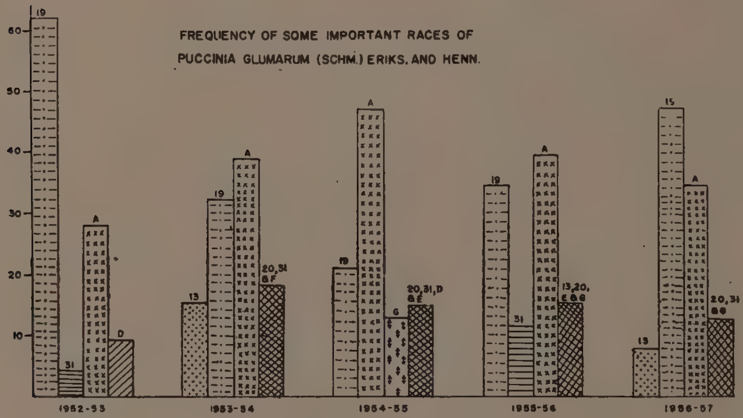
I. Black rust.

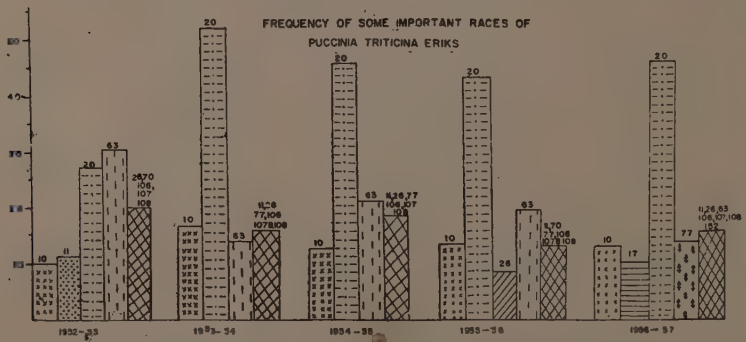
Race	Little club	Marquis	Reliance	Kota	Arnautka	Mindum	Spelmar	Kubanka	Acme	Einkorn	Vernal	Khapli	Reference
17	4	4	0;	4	4	4	4	4	3–4	3–4	0;–1	0;–1	Vasudeva *et. al* (1957)
72	x	0;–2	0;	0;–2	0;	0;–2	0;–2	4	4	3–4	0;–2	4	Vasudeva *et. al.* (1953)
122	3	4	0;	4	4	4	4	4		3	1	1	Gokhale, and Patil, (1952)

II. Brown Rust

Race	Malakof	Carina	Brevit	Webster	Loros	Mediterranean	Hussar	Democrat	Reference
17	4	0;–2	0;–2	0;–1	0;–2	0;–1	4	0;–1	Joshi, *et. al* (1960)
70	2–4	3–4	3–4	3	3–4	0;–1	0;–2	0;–2	Vasudeva *et al* (1957)
77	4	4	4	4	4	4	4	4	Vasudeva *et al* (1955)
162	0;–2	4	4	3	4	4	x	4	Misra, *et al* (1960)







Besides such shifts in occurrence of races, which have a direct bearing on breeding for rust resistance, the appearance of new races is equally disturbing. Very few new races have been isolated during 1952-57 nevertheless, one of them, viz. race 122, appears to be a problem-race

TABLE II-A. Occurrence of Physiologic Races of *Puccinia graminis tritici* (Eriks. & Henn. met with in different states during 1952-57.

Name of State	Wheat or Barley	1952–53 No. of stations	1952–53 No. of samples	1952–53 Races	1953–54 No. of stations	1953–54 No. of samples	1953–54 Races	1954–55 No. of stations	1954–55 No. of samples	1954–55 Races	1955–56 No. of stations	1955–56 No. of samples	1955–56 Races	1956–57 No. of stations	1956–57 No. of samples	1956–57 Races
Jammu & Kashmir	W.	—	—	—	11	12	21, 34, 40 & 42	—	—	—	7	9	21, 40 & 42-B	—	—	—
Punjab and Himachal Pradesh	W.	11	26	21, 34, 40 42 & 194	30	47	15, 21, 34, 40, 42, 122 & 17**.	15	21	21, 34, 40 & 42	9	14	21, 34, 40 & 42	7	25	21, 24, 34, 40 & 42-B
	B.	2	3	21	2	2	21	—	—	—	—	—	—	1	1	21
Delhi	W.	1	6	21, 34, 40 42 & 194	1	11	21, 34, 40 42 & 122	1	3	21, 40 & 42	—	—	—	1	1	42-B
	B.	—	—	—	—	—	—	—	—	—	—	—	—	1	1	42-B
Uttar Pradesh	W.	14	24	21, 34, 40 & 42	17	44	21, 34, 40 & 42	15	20	21, 34, 40 & 42	27	23	15, 21, 34 40, 42 & 42-B	19	43	15, 21, 24, 34, 40 & 42-B
	B.	3	3	21 & 34	2	2	21 & 34	1	1	34	3	3	21 & 40	1	2	15 & 40
Bihar	W.	3	7	21 & 40	4	11	21, 34, 40 & 42	3	11	21, 24 & 42	2	12	21 & 42-B	9	12	15, 21, 34, 40, 42 & 42-B
	B.	1	1	34	4	5	21 & 34	1	1	21	1	2	34 & 42-B	1	2	42 & 42-B
Orissa	W.	—	—	—	1	1	34	1	4	24, 40 & 42	1	1	21	—	—	—
Assam	W.	2	2	21 & 34	—	—	—	1	1	40	1	1	21	—	—	—
Bengal	W.	1	1	21 & 40	—	—	—	—	—	—	—	—	—	2	3	21 & 42-B
Rajasthan	W.	1	1	40	1	1	40	3	3	21 & 42	—	—	—	4	9	21, 40 & 42-B
	B.	—	—	—	1	1	42	—	—	—	—	—	—	—	—	—
Madhya Pradesh	W.	5	7	21, 34, 40 & 42	8	17	21, 34, 40 & 42	5	12	21, 24, 34, 40 & 42	9	21	21, 34, 40 & 42-B	15	29	15, 21, 40 & 42-B
	B.	—	—	—	—	—	—	—	—	—	—	—	—	1	1	42-B
Bombay including Saurashtra	W.	4	4	21, 40 & 42	7	17	15, 21, 34, 40 & 42	17	25	21, 24, 34 40 & 42	32	36	21, 40, 42 & 42-B	37	44	15, 17, 21, 21-A**. 21-A-,1,** 24, 34, 40, 42-B & 122
	B.	—	—	—	—	—	—	1	1	42	—	—	—	1	1	21
Andhra	W.	1	2	21	1	1	34	4	4	21, 24 & 42	4	5	21, 34, 40 & 42	5	5	15, 21, 40 & 42-B
Madras	W.	2	7	21, 34, 40 C* (=72)	4	7	21, 34, 40 & 42	2	3	21	2	4	21, 40, 42 & 42-B	1	9	15, 21, 21- A-1, 34 & 42 B
	B.	—	—	—	—	—	—	2	2	40 & 42	—	—	—	—	—	—
Mysore	W.	6	6	21, 34, 40 & 42	—	—	—	8	12	21, 24, 34, & 42	23	37	15, 21, 34, 40, 42 & 42-B	8	10	21, 40, & 42-B
	B.	—	—	—	—	—	—	—	—	—	—	—	—	1	1	15
Kerala	W.	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Total No. of Wheat Samples:		93				169			119			163			190	
Total No. of Barley Samples:		7				10			5			6			10	

** New races

Total Number of Samples: Wheat — 734 and Barley = 38

TABLE II-B. Occurrence of Physiologic Races of *Puccinia triticina* Eriks. in different States during 1952-57.

Name of State	1952–53 No. of stations	1952–53 No. of samples	1952–53 Races	1953–54 No. of stations	1953–54 No. of samples	1953–54 Races	1954–55 No. of stations	1954–55 No. of samples	1954–55 Races	1955–56 No. of stations	1955–56 No. of samples	1955–56 Races	1956–57 No. of stations	1956–57 No. of samples	1956–57 Races
Jammu & Kashmir	—	—	—	1	3	20	—	—	—	1	1	20	—	—	—
Punjab & Himachal Pradesh	10	18	10, 11, 20, 26, 63, 70,** 106 & 107	6	16	10, 11, 20, 26, 63, & 77**	14	36	10, 11, 20, 26 & 63	7	27	10, 20, 63, 70, 77 & 107	2	14	10, 20 & 63
Delhi	1	2	20 & 63	1	9	10, 11, 20 & 108	2	5	10, 20 & 63	3	8	10, 20 & 63	—	—	—
Uttar Pradesh	13	16	11, 20, 63, 107 & 108	23	74	10, 20, 26, 63, 77**, 107 & 108	17	48	10, 20, 63, 77, 107 & 108	26	58	10, 11, 20, 26, 63, 70, 106, 107 & 108	20	49	10, 11, 17** 20, 26, 63, 77, 106, 107 & 162
Bihar	2	6	20, 63, 106, & 107	7	29	10, 20, 63 & 77**	3	20	10, 20, 63, 77, 107 & 108	3	20	10, 20, 26, 63, 77 & 107	9	20	10, 17** 20 & 77
Orissa	1	2	63 & 108	3	3	11, 20 & 63	1	3	20 & 63	4	10	10, 20, 26, 63 & 106	3	3	20 & 107
Assam	—	—	—	—	—	—	—	—	—	2	2	20	—	—	—
Bengal	4	7	20, 63 & 106	—	—	—	3	17	10, 20 & 63	2	5	10, 20 & 106	3	5	10, 20, 63 & 106
Rajasthan	—	—	—	6	17	10, 20, 63 & 107	1	1	20	—	—	—	3	8	20, 63 & 107
Madhya Pradesh	2	3	10, 20, 63 & 106	3	5	10, 11, 20 & 107	2	4	10 & 20	9	14	10, 11, 20, 63, 77, 106 & 162**	9	15	10, 17** 20, 63, 77 & 107
Bombay (including Saurashtra)	3	3	10, 11, 20 & 63	8	16	10, 20, 26, 63, 77, 107 & 108	9	22	10, 20, 26, 77, 107 & 108	12	16	10, 20, 26 & 107	22	30	10, 17**, 20, 77 & 108
Andhra	1	2	63	1	1	10	2	6	20 & 26	—	—	—	5	8	17 **, 20, 26, 63, 77 & 107
Madras	7	10	11, 20, 63, 106, 107 & 108	1	7	10, 20, 63, & 107	6	10	10, 20 & 63	2	6	20, 26, 63, 77 & 107	3	16	10, 11, 17**, 20, 26 63, 77 & & 108
Mysore	8	8	10, 11, 20 & 63	4	6	10, 20, 26, 106, 107 & 108	3	4	20, 63, & 107	3	5	20, 63 & 107	2	2	77
Kerala	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Total No. of Wheat Samples		77			186			176			172			170	

Total Number of Samples: 781.

** New Races

Table II-C. Occurrence of Physiologic Races of *Puccinia glumarum* (Schw.) Erikss. & Henn. in different States during 1952-57.

Name of State	1952–53 No. of stations	1952–53 No. of samples	1952–53 Races	1953–54 No. of stations	1953–54 No. of samples	1953–54 Races	1954–55 No. of stations	1954–55 No. of samples	1954–55 Races	1955–56 No. of stations	1955–56 No. of samples	1955–56 Races	1956–57 No. of stations	1956–57 No. of samples	1956–57 Races
Jammu & Kashmir	—	—	—	—	—	—	—	—	—	1	1	A	—	—	—
Punjab & Himachal Pradesh	9	16	19, 31, A & D	9	20	13, 19, 20 & A.	13	24	19, 20, 31, A, D & E	12	35	13, 19, 20, 31, A & D	14	32	13, 19, 31 & A
	2	2*	19 & A	—	—	—	2	2*	19	4	6*	19 & 31	1	1*	19
Delhi	1	1	19	1	2	A & F	—	—	—	1	1	A	1	1	19
Uttar Pradesh	5	5	19 & D	3	9	13, 19, 31 & A	5	7	19 & A	7	14	19, 31 & A	10	16	19, 31 & A
	4	4*	19	—	—	—	2	2*	G	3	4*	19 & G	2	2*	19
Bihar	2	2	19 & D	1	1	19	—	—	—	—	—	—	2	3	13, 20 & A
Orissa	—	—	—	—	—	—	1	1	A	—	—	—	—	—	—
Assam	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Bengal	1	1	19	—	—	—	1	2	A	1	1	19	—	—	—
Rajasthan	—	—	—	2	2	19 & A	—	—	—	—	—	—	—	—	—
Madhya Pradesh	—	—	—	—	—	—	—	—	—	3	3	19 & A	2	3	13 & A
Bombay (including Saurashtra)	—	—	—	1	1	19	—	—	—	—	—	—	—	—	—
Andhra	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Madras	—	—	—	—	—	—	—	—	—	—	—	—	1	2	19 & A
	1	1*	G	—	—	—	1	3*	G	1	1*	G	1	3*	G
Mysore	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Kerala	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Total Number of wheat samples		25			35			34			55			57	
Total Number of Barley samples		7			—			7			11			6	

Total Number of Samples: Wheat — 206 Barley = 31

* Indicating Barley Samples.

TABLE III. Frequency of occurrence of races *Puccinia graminis tritici*

Year of collection	Total No. of samples analysed	Total No. of isolates	Frequency of in percentage															
			15	15-C	17	21	21-A	21-A-1	24	34	40	42	42-B	72	75	117	122	194
1952–53	100	122	—	—	—	45.91	—	—	—	9.83	26.23	15.58	—	—	—	—	—	—
1953–54	179	212	1.89	—	0.47	49.06	—	—	—	9.94	19.34	18.40	—	—	—	—	0.90	—
1954–55	124	125	—	—	—	35.56	—	—	8.16	15.56	15.56	15.16	—	—	—	—	—	—
1955–56	169	186	1.06	—	—	30.66	—	—	—	8.60	12.38	21.50	25.80	—	—	—	—	—
1956–57	200	222	4.95	—	0.45	29.28*	0.45	1.35	1.35	7.66	12.62	0.45	40.99	—	—	—	0.45	—

* Identification of biotypes 21-A and 21-A-1 was taken up only after 1956.
Identification of biotype 42-B was taken up only after 1955.

Puccinia triticina

Year of collection	Total No. of samples analysed	Total number of isolates.	Frequency of races in percentage											
			10	11	17	20	26	63	70	77	106	107	108	162
1952–53	77	110	10.00	11.83	—	27.27	2.75	30.91	0.91	—	5.45	6.36	4.52	—
1953–54	186	209	16.75	2.39	—	52.63	4.32	14.36	—	2.39	0.47	4.30	2.39	—
1954–55	176	302	12.38	2.48	—	46.04	4.45	22.28	—	3.95	1.98	4.95	1.49	—
1955–56	172	186	13.90	1.62	—	44.08	8.60	19.90	0.54	3.22	3.22	4.30	0.54	—
1956–57	170	181	13.25	1.10	9.96	46.98	2.20	6.09	—	13.27	1.10	4.40	1.10	0.55

Puccinia glumarum

Year of collection	Total No. of samples analysed	Total number of isolates	Frequency of races in percentage									
			13	19	20	31	A	D	E	F	G	H
1952–53	32	33	—	60.60	—	3.04	27.27	9.09	—	—	—	—
1953–54	35	35	14.28	31.42	8.57	5.75	37.13	—	—	2.85	—	—
1954–55	41	41	—	26.86	4.87	4.87	46.35	2.43	2.43	—	12.19	—
1955–56	66	70	7.14	34.29	1.42	11.43	38.57	—	4.29	—	2.86	—
1956–57	63	63	7.93	46.05	1.58	6.35	33.33	—	—	—	4.76	—

on account of the fact that most of our improved varieties are susceptible to it under glasshouse conditions. Another race that is equally dangerous to some of the improved wheat varieties is race 77 of brown rust. This race was first picked up in 1954 from Pusa (Bihar) on the Mediterranean Variety, a brown rust differential, which was resistant to all the then prevailing races in India. The race has now been recorded from other place in the plains.

As mentioned earlier, for the study of physiologic races of black rust, a set of 12 differential host originally selected by Stakman and Levine (1922), have been used. It has, however, been observed by various workers that rust collections which show identical reactions on differential varieties, at times gave appreciably different reactions on some other varieties. The diversity of reactions may have been due to environmental conditions or to different biotypes. Waterhouse and Watson (1941) have shown that race 34 of black rust in Australia is different in its host-range and pathogenicity from race 34 of the United States.

In India, Uppal and Gokhale (1947) and Gokhale (1950) identified two new biotypes of race 42 and another of race 15 by using wheat varieties Yalta and Charter. Recently Prasada and Sreekantiah (1956) have reported occurrence of a biotype in race 21 and designated it as 21-A.

It has been pointed out by Stakman (1954) that a large number of biotypes may be present in each race and that the number of such biotypes may vary with change of varieties used for the purpose. The occurrence of biotypes makes the task of breeding for resistance still more difficult as some of the important varieties found resistant to the original race may succumb to its biotypes. In this respect the biotypes of races 15 and 42 have been found to be more virulent on some of the important wheat varieties than the races themselves as shown by the data set out in table IV.

TABLE IV.

Reaction of some wheat varieties to races 15 and 42 of black rust and their biotypes.

Names of varieties	15	15–C	42	42–B
E. 124	—	—	0	4
E. 569	—	—	2	2–3
E. 1914	1	4	—	—
NP 798	0	3–4	—	—
NP 799	0	4	—	—
NP 807	—	—	2	4
NP 809	0;–1	3	—	—

No precise reason for the sporadic appearance of new races has been given so far. It is believed that the so-called new races might have existed in the past but their presence could not be detected due to their restricted

distribution and inadequate sampling. Some of the newly isolated races like 72 and 122 of black rust have so far been isolated only rarely. The possibility of introduction of new races from foreign countries cannot, however, be ruled out. Although, Himalayas in the North and Indian Ocean in the South would, under normal circumstances, be effective barriers for air-borne dissemination of rust spores, their introduction to a limited extent is possible through the agency of aerial intercontinental communications or from neighbouring countries like Afghanistan and Iran.

Mutations in cereal rusts are by no means rare but in India so far no case of a mutation involving changes in pathogenicity has been recorded although pure cultures of wheat rust races have been maintained for nearly 30 years on a susceptible variety. Yet another possibility of the appearance of new races is through hybridization on *Berberis* in some remote parts of the mountainous regions of the country (Vasudevan—1954). Unlike temperate countries, the teleutospores in India represent the summer stage in the life cycle of these rusts. The teleutospores in the plains are formed after the month of March and are rendered inviable due to the high temperatures that prevail in subsequent months. So that even if disseminated by winds to the hills they cannot infect susceptible *Berberis* species like *B. lycium*. In the hills, the teleutospores are formed in May or June depending on the altitude of cultivation. Taking into consideration the susceptibility of certain indigenous specei:s of *Berberis* e.g. *B. lycium* and also the possibility of occurrence of *Berberis vulgaris* in the interior of the hills, the chances of infection of *Berberis* at at higher altitude cannot altogether be ruled out.

It was primarily with the object of locating pockets of barberry infections that regular survey of Lahaul Valley (10,000–ft. asl.) in the Himalayas, where wheat is grown as a summer crop (May – September) was carried out. Judging from the time of sowing of wheat, the temperature prevailing at the time of formation of teleutospores and their subsequent exposure to snow, the presence of a species of *Berberis* and finally the abundance of moisture in spring and early summer, it was felt that *Berberis* might play the part of functional alternate host in this area like most of the temperate countries. Visits to this Valley were made in May and June 1954 and again in June-July 1955. From the data collected so far it was observed that:

(i) The *Berberis* species occurring in Lahaul Valley is *B. Jaeschkeana* which is resistant to black rust under laboratory conditions.

(ii) The aeciospores collected from *Berberis* did not infect wheat though they infected an unidentified grass.

(iii) The races occurring in Lahaul Valley are mostly the same as in other parts of the country.

It is indicated, therefore, that *Berberis* sp. in Lahaul Valley is not connected with wheat rust. Intensive search for barberries infected with wheat stem rust is, however, being continued in the hills.

Another possibility of appearance of new races is through heterokaryosis (Nelson, Wilcoxson and Christensen, 1955) or *somatic hydbridization* (Watson 1957). Work on those lines is necessary to establish if those phenomena are of frequent occurrence under Indian conditions and would thereby account for the origin of new races in nature.

Summary

1. For the identification of physiologic races, 1,721 collections of black, brown and yellow rusts of wheat and 69 collections of black and yellow rusts of barley, obtained during the period 1952 to 1957 from those crops from all over the country were studied on standard differential hosts. Races 15, 17, 21, 21-A, 21-A-1, 24, 34, 40, 42, 42-B, 72 and 122 of black rust; races 10, 11, 17, 20, 26, 63, 70, 77, 106, 107, 108 and 162 of brown rusts and races 13, 19, 20, 31, A, D, E, F and G of yellow rust were identified. All the races of yellow rust had been met with before but in the case of black and brown rusts, races 17, 72, 122, 21-A, 21-A-1 of the former and races 17, 70, 77 and 162 of the latter were located during 1952-57.

2. Throughout this period, race 21 of black rust was most predominant. Other important races were 34, 40 42, and 42-B. In the case of brown rust, race 20 was most wide-spread followed by race 63. Races A and 19 of yellow rust continued to be most important.

3. No evidence of barberies functioning as alternate host of black rust could be obtained.

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STUDIES ON A MOSAIC DISEASE OF COWPEA (VIGNA SINENSIS SAVI)

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Introduction: A mosaic disease of cowpea (*Vigna sinensis* Savi) was observed in the farm area of this Institute during October 1958. The incidence of the disease was found to be as high as 50 per cent in some of the varieties. The disease is characterised by mosaic mottling of the leaves accompanied by distortion and reduction in leaf size. The younger leaves produced at the growing point show vein clearing followed by mottling and slight blistering (Fig. 1). The older leaves are generally chlorotic and pale. The infected plants bear only a few pods. The pods from infected vines are usually small and shrunken, containing only a few shrivelled seeds which in most cases are not viable. The yield is considerably reduced. The investigations conducted on the mode of transmission, host range and physical properties of the causal virus are reported in this paper.

Material and Methods: All experiments were conducted in an insect-proof glass house. The culture of the virus was obtained from naturally infected cowpea plants and was maintained by successive transfers by mechanical inoculation to healthy seedlings. Mechanical inoculation was made by the leaf rubbing method using carborundum powder as an abrasive. The standard extract of the virus was prepared by crushing young infected leaves to a fine pulp adding distilled water at the rate of one c.c. per gram of leaf material. For insect transmission tests healthy colonies of insects maintained in an insectary were used.

Transmission of the Disease: Mechanical inoculation: The disease was successfully transmitted to healthy cowpea plants by mechanical inoculation with the expressed juice from the mosaic affected plants by the leaf rubbing method using carborundum powder as an abrasive. The symptoms appeared on the inoculated plants in about 7–10 days after inoculation in the form of vein clearing followed by mosaic mottling of the leaves (Fig. 2).

Insect Transmission: Four species of aphids viz. *Aphis craccivora* Koch., *Aphis evonymi* Fabr., *Aphis gossypii* Glov. and *Myzus persicae* Sulz. were tested in an attempt to determine the insect vector of the cowpea mosaic virus. The aphids were starved for 2 hours and given an acquisition feeding period of 30 minutes before removing them to healthy cowpea plants. They were allowed to feed for 24 hours on the test plants after which they were killed by spraying the plants with 0.1 per cent Ekatox. Successful transmission of the disease was obtained with all the four species.

SEED TRANSMISSION: The seed collected from mosaic infected cowpea plants from the field as well as from artificially infected plants in the glass house were sown in 6" pots in the insect-proof house and observations were recorded on the seedlings, by counting the number of seedlings showing mosaic symptoms. It was observed that the cowpea mosaic virus is transmitted through the seeds of mosaic affected plants and the seed transmission in different samples of seeds collected varied from about 4 to 40 per cent. The symptoms of the disease usually appeared on the first trifoliate leaf (fig. 3) but occasionally even the primary simple leaves showed mosaic mottling.

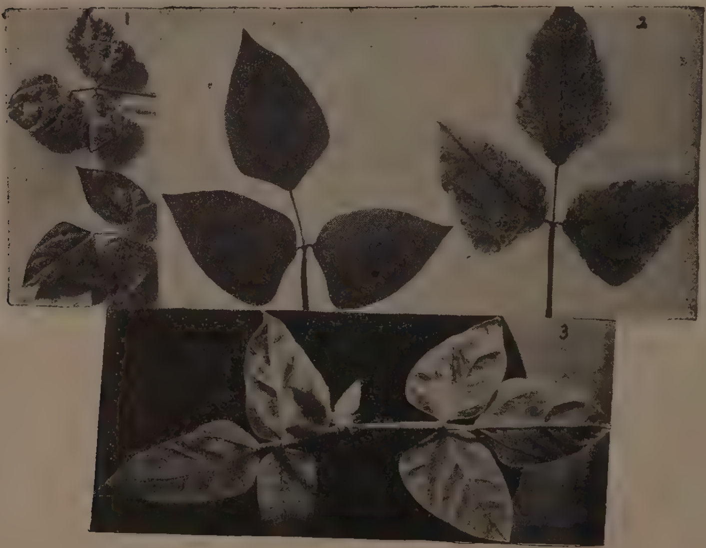

Fig. 1. Healthy and mosaic affected leaves of cowpea under field conditions.

Fig. 2. Healthy and infected leaves of cowpea plants inoculated by mechanical inoculation under glass house conditions.

Fig. 3. Transmission of the cowpea mosaic virus through seed of infected plants.

HOST RANGE: Different species and varieties of plants belonging to 13 families were inoculated with the infective virus sap by the usual leaf rubbing method using carborundum powder as an abrasive. The inoculated plants were kept under observation for 4 to 5 weeks for the appearance of symptoms. The plant species which did not exhibit any visible symptoms of the disease after this period were indexed on healthy cowpea seedlings to determine if they were carrying the virus symptomlessly. This was done by macerating the leaves from the apparently healthy inoculated plant species in a pestle and mortar and inoculating the sap on healthy cowpea seedlings.

It was observed that the virus is transmissible to *Vigna sesquipedalis, V. cylindrica, V. nilotica, V. putigera, V. vexillata, Phaseolus aureus, P. mungo, P. lathyroides, P. limensis, P. lunatus* and *Canavalia ensiformis*, (Figs. 4–7) all belonging to the family Leguminosae. Systemic mosaic symptoms were produced in all the above plant species except *Vigna vexillata* which produced local necrotic lesions on the inoculated leaves only (Fig. 8). None of the other plant species that were tested was infected nor any of them proved to be a symptomless carrier of the virus.

Fig. 4. Leaf of *Vigna sesquipedalis* infected with cowpea mosaic virus.
Fig. 5. Leaf of *Vigna cylindrica* infected with cowpea mosaic virus.
Fig. 6. Leaf of *Phaseolus mungo* infected with cowpea mosaic virus.
Fig. 7. Leaf of *Canavalia ensiformis* infected with cowpea mosaic virus.

Physical Properties: The standard extract of the virus was found to be infective when heated to 95 °C for 10 minutes but was rendered innocuous when heated to 98.5 °C for the same period. It was infective after storage for 12 days at room temperature (13–19 °C) but was rendered noninfective after 13 days. At the frigidaire temperature (8-10 °C) the virus could withstand storage for 20 days but not for 21 days. The infective leaf extract when diluted to 1 : 30,000 with water was still infective but was rendered non-infective when diluted to 1 : 40,000. The virus was found to be viable in desiccated mosaic infected cowpea leaves for 5 days at room temperature (30–43 °C) but was inactivated after 6 days. The

Fig. 8. Inoculated leaf of *Vigna vexillata* showing local necrotic lessions.

virus in the undiluted leaf extract was not completely inactivated even after exposure of 3 hours (240 m.u.d.)* to the ultraviolet light irradiated from a distance of 24 inches by an Alpine Sunlamp Model IX 220 Volts DC., although the infectivity was very much reduced. In the virus extracts diluted to 1 : 10, 1 : 100, 1 : 1000 and 1 : 10,000 the infectivity was lost after an exposure for 3, 2, 1 and ½ an hour respectively.

pH Stability: For the study of pH stability range of cowpea mosaic virus, the method described by Samuel, Best and Bald (1935) was followed, using composite buffer solution of 0.0533 M with respect to each of boric acid, potassium pthalate, and potassium dihydrogen phosphate. Samples of the buffer solutions were adjusted to different pH values ranging from 1.1 to 12.1 by the addition of 0.2 M sodium hydroxide or 0.2 M hydrochloric acid and finally diluted with water to give a final solution of the desired pH and being 0.04 M with respect to the total buffering substances. One c.c. of centrifuged virus extract was added to 9 c.c. of different adjusted solutions and mixed well. The inoculations were made with these samples adjusted to different pH values immediately after preparation as well as after storage for 5 to 10 days at frigidaire and room temperatures.

It was observed that the cowpea mosaic virus can withstand a very wide range of H-ion concentration i.e. pH 1.1 to 12.1 when inoculations were made immediately after adjustment of the pH. The pH range, however, was found to be reduced to between 2.0 and 9.1 when the pH adjusted samples were stored at room temperature (25–36 °C) for 10 days. At the frigidaire temperature (8–10 °C), the virus remained infective over pH range of 1.1 to 12.1 although its infectivity was reduced after storage as judged by the low percentage of infections obtained.

Effect of Chemicals: Double strength standard extract of the virus was prepared and centrifuged at 3,000 r.p.m. for 30 minutes in a

*A minimal unit of dosage (m.u.d.) of exposure to ultraviolet irradiation is 5,22,000 ergs per sq. cm. of Erythema producing ultraviolet rays.

"Rotofix 2,800" centrifuge. To the supernatant were added double strength solutions of the chemicals to be tested in equal proportions. In this manner the virus in standard strength extract was subjected to the action of the desired strength of the chemical. Immediate inoculations were made on cowpea plants followed by inoculations after 1 hour and 24 hours after storage of the mixtures in a frigidaire (8–10°C).

The results of these tests showed that the cowpea mosaic virus can withstand treatment with 40 per cent alcohol, 50 per cent hydrogen peroxide, 0.25 per cent mercuric chloride and 0.5 per cent potassium permanganate even after 24 hours' reaction with the chemicals. The virus was, however, inactivated by 1 per cent potassium permanganate, 0.5 per cent mercuric chloride and 0.5 per cent formalin after reaction with the chemicals for 24 hours and by 1–2 per cent formalin even after one hour.

Discussion: Studies on mosaic diseases of *Vigna* spp. and their causal viruses have been conducted by McLean (1941), Snyder (1942), Yu (1946), Dale (1949), Anderson (1955) and Capoor *et al*. (1947, 1956). Mosaic disease of *Vigna unguiculata* and *Vigna catjang* have also been reported from India by Thomas (1937) and Vasudeva (1942) respectively. In addition cowpea mosaic viruses have been reported by Oliveira (1947) and Warid and Plakidas (1950, 1952). The mosaic virus reported by Dale (1949) is transmitted by the leaf beetle, *Ceratoma ruficornis* and is distinct from the others which are aphid transmitted. The viruses reported by McLean (1941), Yu(1946), Snyder (1942) and Anderson (1955) resemble each other and have been designated as *Marmor vignae*. Capoor *et al* (1956) distinguished their virus on *Vigna cylindrica* as a strain of *Marmor vignae* on account of very high thermal death point of the virus and designated it as *Marmor vignae* var. *catjang*.

The virus under study, resembles closely the mosaic virus (*Marmor vignae* var. *catjang*) reported by Capoor *et al* (1947, 1956) in physical properties, host range, seed transmission and in having aphids as its vectors. In addition to the aphid vectors reported by the authors, *Aphis evonymi* has been found to be an additional vector. *Vigna nilotica*, *V. putigern*, *Phaseolus aureus*, *P. mungo*, *P. lathyroides* and *P. limensis* proved to be additional hosts of the virus and the virus induced local necrotic lesions on the inoculated leaves of *Vigna vexillata*.

Summary

A mosaic disease of cowpea (*Vigna sinensis* Savi) characterised by mosaic mottling of the leaves accompanied by blistering and distortion has been described.

The causal virus is sap transmissible and is carried in seed obtained from diseased plants. *Aphis craccivora* Koch., *A. gossypii* Glov., *A. evonymi* Fabr., and *Myzus persicae* Sulz. have been established to be the vectors of the virus. The host range of the virus is restricted to the family Leguminosae. The virus induces local necrotic lesions on *Vigna vexillata*.

The physical properties of the virus which has been identified as *Marmor vignae* var. *catjang* have been described.

ACKNOWLEDGEMENT: The authors are extremely grateful to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his keen interest and valuable suggestions throughout the course of these investigations.

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STUDIES ON THE SURVIVAL OF PLANT PATHOGENS ADDED TO THE SOIL 1. FUSARIUM SPP. AND XANTHOMONAS CASSIAE

G. Rangaswami and N. N. Prasad

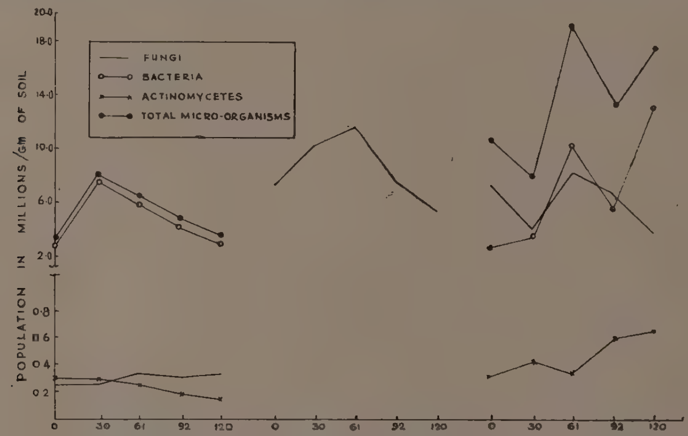
(Accepted for publication March 15, 1961)

Soil-borne plant pathogens are a great menace to the agriculturists. The survival and persistence of these organisms in the soils is one of the baffling problems engaging the attention of microbiologists for several decades now. The fate of the pathogens when added to the soil has been investigated in some cases, but more detailed investigations are required to understand the complicated problem. It is generally understood that certain fungi and bacteria die out in a short period when added to the soil while others live for several years. The main reason for this is perhaps the limitations in the availability of specific food requirements of the organism. But soil is a complex biological system wherein a great mixture of microbial population exists. The interrelationship of these organisms are so formulated that a biological equilibrium is maintained in the soil. According to Waksman (1956) all attempts made to isolate specific antibiotic substances or to demonstrate their presence in the soil have so far failed and there is no justification to maintain that antibiotics play a definite role in maintaining the balance between living organisms of the soil. The plant pathogens reaching the soil are subject to various physical, chemical and biological influences of the soil. If one can understand at least some of these factors acting on the pathogen it might be possible to advantageously utilize the knowledge to check the spread of pathogen in the soil. With this view in mind some plant pathogens were added to the soil and the changes taking place in the biological system of the soil studied. The results are presented here.

Material and Methods: Two species of *Fusarium*, one causing pre-emergence rot of wheat seeds and the other causing pre- and post-emergence rot of ragi seeds (*Eleusine coracana* Gaertn.) and a bacterium, *Xanthomonas cassiae* Kulkarni *et al*, causing post-emergence rot of *Cicer arietinum* L. were studied by the soil enrichment technique. The fungi and the bacterium were multiplied in sand-oat meal medium in 250 ml. Erlenmeyer flasks for 10 days. The soil used in these studies was loamy and was collected from the fields of the Annamalai University Experimental Farm. It was filled in one lb. wide-mouthed jam bottle upto three-fourths of the volume, closed with a wooden lid-cotton wool plug and sterilized by autoclaving at 30 lbs. pressure for two hours. The organisms grown in sand-oat meal medium were added to the sterilized and unsterilized soils in the proportion of 10 : 1 of the soil and the inoculum. On inoculation the bottles were incubated at room temperature (28–30°C). Samples from each of the soils were analysed at periodical intervals for the microbiological population by the dilution plate method using Ken Knight's agar, yeast extract agar and potato dextrose agar media. The average

number of fungi, bacteria and actinomycetes present at periodical intervals in one gram of the soil, on a dry weight basis, was calculated.

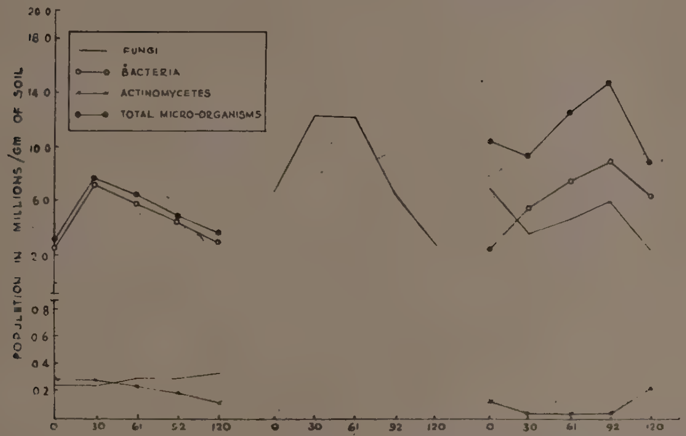
EXPERIMENTAL RESULTS; 1. *Fusarium sp. from Wheat:* When the fungus was added to the sterilized soil, there was an increase in the population of soil, which later on declined slowly. When it was added to unsterile soil there was an increase in the total fungal population of the soil, which got reduced after an interval of 60 days. Subsequent to the addition of the *Fusarium* there was an increase in the actinomycete population which was maintained at high level even after 120 days. The bacterial population increased considerably after 60 days, along with a rapid reduction in the fungal population (Fig. 1).



INCUBATION IN DAYS UNAMENDED SOIL | INCUBATION IN DAYS STERILE SOIL + WHEAT FUSARIUM | INCUBATION IN DAYS UNSTERILE SOIL + WHEAT FUSARIUM

2. *Fusarium sp. from Ragi:* When the fungus was added to the sterilized soil there was an increase in the population, which after 60 days started declining rather abruptly. When it was inoculated into unsterile soil there was a decline in the fungal population, whereas the bacterial population was steadily on the increase upto 90 days. There was a reduction in the actinomycete population in the initial stages, which increased after 60 days. In general, all the organisms in the soil got reduced in population after about 90 days (Fig. 2).

3. *Xanthomonas cassiae:* When the bacterium was added to sterile soil, there was rapid increase in cells upto 30 days after which there was reduction. But when added to the unsterile soil the bacterial population gradually increased upto 60 days, where after there was a decline, the population reaching the level of that of unamended soil. The fungal population also behaved almost in a similar manner, except for a rapid reduction after 90 days. The actinomycete population, on the other hand, showed

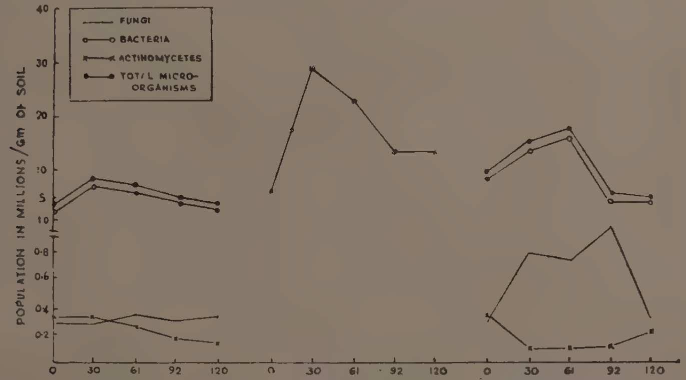


INCUBATION IN DAYS UNAMENDED SOIL

INCUBATION IN DAYS STERILE SOIL+RAGI FUSARIUM

INCUBATION IN DAYS UNSTERILE SOIL+RAGI FUSARIUM

a decrease in the initial stages, after which there was a slight, but steady, increase in the population upto 120 days (Fig. 3).



INCUBATION IN DAYS UNAMENDED SOIL

INCUBATION IN DAYS STERILE SOIL+ X. CASSIAE

INCUBATION IN DAYS UNSTERILE SOIL+ X. CASSIAE

Discussion: Both the fungi and the bacterium were found capable of establishing and multiplying in the soil as indicated by the results obtained with the sterile soil. The reduction in the population of the added organisms in sterile soil after some time might be due to exhaustion of the food material. There has been fluctuations in the microbial population of unamended (control) soil, when estimated at different intervals,

which might be due to exhaustion of food material or due to other external factors, which are common to all the treatments in the experiment (Figs. 1 to 3). When the *Fusarium* spp. from wheat and ragi were added to the unsterilized soil there has been a decline in the fungal population, as compared to that in the sterilized soil, indicating thereby that the fungi were not quite free to establish and multiply in the soil. This decline seems to be accompanied by an increase in bacterial population and to some extent by the actinomycete population. The increase in the populations of these two types of organisms seems to be rather significant when compared to their gradual reduction in the unamended (control) soil. In the case of X. *cassiae* the organism was found to establish and slowly multiply in the unsterile soil but soon found to decline. This decline was accompanied by an increase in the fungal population, which was nearly three times that of the fungal population in unamended (control) soil. There was also a slight increase in the actinomycete population.

The results obtained suggest that the micro-organisms normally present in the soil influence considerably the establishment, multiplication and survival of the added plant pathogens, which might otherwise readily establish and multiply in the soil. Bacteria, and to some extent actinomycetes, seem to play a prominent role in reducing the *Fusarium* spp. added to the soil, whereas fungi, and to a limited extent actinomycetes, seem to play a role in reducing the population of *X. cassiae* added to the soil. The possibilities of soil-dwelling saprophytes acting on the added pathogen and reducing the pathogenicity have been studied with *Trichoderma* spp. by Allen and Haenseler (1935), Christensen (1936) and Weindling and Fawcett (1936). In the present investigations, though no particular attention was paid to the genera or species of micro-organisms that are acting against the added pathogens, it is quite probable that some of the antagonistic bacteria, fungi and actinomycetes play a role in suppressing the growth of the added pathogens.

SUMMARY

With a view to study the survival of plant pathogens in the soil, *Fusarium* sp. causing pre-emergence rot of wheat seeds, *Fusarium* sp. causing pre- and post-emergence rot of ragi seeds and *Xanthomonas cassiae* Kulkarni *et al* causing post-emergence rot of *Cicer arietinum* L. were added to both sterilized and usterilized soils and the changes in the microbial population of the soil were estimated at periodical intervals upto 120 days. Though the pathogens were found capable of establishing and multiplying in the sterile soil, they were found to be suppressed when added to unsterile soil. The suppression of the two fungi was accompanied by increases in the bacterial and actinomycete populations of the soil and that of the bacterium was accompanied by increase in the fungal and actinomycete populations of the soil indicating thereby the possible inhibitory effect of these organisms on the added pathogens.

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APPLICATION OF FUNGICIDES IN THE CONTROL OF SECONDARY AIR-BORNE INFECTION OF HELMINTHOSPORIUM ORYZAE BREDA DE HAAN

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(Accepted for publication May 10, 1961)

Paddy (*Oryza sativa* L.), in West Bengal suffers every year from the attack of brown spot of *Helminthosporium oryzae* Breda de Haan. The disease is evident from the seedling stage when it is manifested by the presence of infected seedlings in the seed bed, arising out of the seeds having primary seed-borne infection (Chattopadhyay, 1951). The infection, which is first evident in the coleoptile, passes to the developing leaves which show the presence of a number of small sized spots.

From the tillering phase, as weather conditions become more favourable for production and dissemination of conidia, the infection gradually progresses and the leaves become progressively spotted with brown ellipsoidal spots which gradually increase in size and eventually the grains are also infected. Under favourable conditions for the spread of the disease, the infection may be severe resulting in serious loss in yield.

In recent years in India, Vaheeduddin (1953), Padmanabhan, Ganguly and Chandwani (1956) reported control of blast disease of paddy by spraying different fungicides. In Japan, direct control of the disease has been achieved by the application of organo-mercurial dust (Padwick, 1956).

In view of the fact that more knowledge on the control of secondary air borne infection of *Helminthosporium oryzae* is desirable a number of experiments were undertaken, the results of which are presented below.

An experiment was conducted on Patnai 23 variety with two different fungicides, namely Perenox and Dithane-Z–78 applied both as spray and as dust. Two applications were given altogether, with variations in the time of application. In one case both the applications were made before flowering–the first one in the 3rd. week of September and the second one in the second week of October. In another case the first application was made before flowering in the 3rd. week of September and the second one in the 1st. week of November, after flowering.

Seeds were treated, one week prior to sowing, with Agrosan GN. The experiment was conducted for two consecutive years in replicated randomized plots with 4 replications and the plot size was 15′ x 12′.

Data taken of leaf infection together with its statistical analysis are presented below. Each figure in Table I represents an average of four replications.

TABLE I. Effect of application of different fungicides on the intensity of infection of *Helminthosporium oryzae* on Patnai 23 variety of paddy.

Treatment	Leaf infection value	
	1952	1953
Control	576.63	107.25
Perenox (spray-4 lbs in 100 gallons of water) 1st type of application	344.50	90.50
Perenox (spray) ... ... 2nd type of application	468.63	103.63
Perelan (dust-20 lbs. per acre) ... 1st type of application	427.25	94.75
Dithane-Z-78 (spray-2 lbs. per 100 gallons of water) ... ... 1st type of application	487.12	92.13
Dithane-Z-78 (spray) ... ... 2nd type of application ...	417.00	90.25
Dithane-Z-78 (dust) @ 20 lbs. per acre ... 1st type of application	467.38	95.50
Dithane-Z-78 (dust) ... ... 2nd type of application	428.50	89.75

Critical difference (between treatments)
at 5% level – 1.28
1% level – 2.34

Critical difference between figures of treatment X year
at 5% level – 1.51
1% level – 2.04

It may be concluded that application of fungicides (spray and dust) results in the reduction of intensity of infection as compared with the control. The magnitude of reduction however varies.

During 1953 a trial was undertaken in cultivators' plots, on a compact block of 33 acres in *Sarisha*, in 24–Parganas. Perenox was sprayed twice at an interval of 15 days between tillering and flowering on two varieties namely 'Jamainaru' (of coarse grain) and 'chamarmani' (of fine grain). Data were taken of the leaf infection, grain infection and yield of grain. For recording leaf and grain infection, 25 plants were selected at random for each variety and from each plant 5 leaves were taken for leaf infection and 5 ear-heads for grain infection. Yield of grain and straw was recorded on the twenty-five plants selected at random. Similar observations were made on the same varieties unsprayed and growing in the adjacent area. Data are presented in table II, each figure representing an average of 25 random samples.

TABLE II. Effect of application of Perenox on the infection and yield of two varieties of paddy.

Name of variety	Treatment	Leaf infection value	Percentage grain infection	Average yield of grain (of 25 plants in lbs
Jamainaru	Unsprayed	45.0	32.3	1.80
	Sprayed	35.4	20.0	2.25
Chamarmani	Unsprayed	42.6	16.6	2.26
	Sprayed	26.4	14.1	2.45

Spraying with Perenox resulted in reduction of both leaf infection as well as grain infection. Increase in yield of grain was also due to the reduced leaf and grain infection.

In recent years organo-mercurial dust is extensively used in Japan for controlling the leaf spot diseases of paddy (Padwick, 1960). To compare the efficacy of organo-mercurial dusts with copper fungicides, an experiment was laid out at State Agricultural Farm, Coochbehar with two varieties of paddy—Dudswar (Aman—periodically fixed) and Dharial (Aus—timely fixed). The fungicides used are stated below:

1. Organo-mercurial dust (containing 1 per cent mercury supplied by Messers Imperial Chemical Industries (India) Private Ltd. (2) Cupravit Blue OB (proprietory copper fungicide product of Messers Bayer & Co., Lever-Kusen, West Germany, and (3) Perenox. Altogether four applications were given after transplanting and before flowering. The experiment was laid out in replicated randomized blocks having five replications for each treatment with plot size 40′ x 12′. Data taken of leaf infection, grain infection, and yield of grain are presented in the following tables.

TABLE III.—Effect of application of fungicide on the control of secondary air borne infection of *Helminthosporium oryzae* and yield of Dudswar variety of paddy.

Treatment	Dose	Leaf infection value	Percentage of infected grain	Yield of grain (in lbs.)
Control		106.0	71.2	12.02
Organo-mercurial dust	20 lbs. per acre	25.2	34.4	15.4
Cupravit Blue OB	4 lbs. per 100 gallons of water	32.6	41.2	14.2
Perenox	4 lbs. per 100 gallons of water	35.3	50.1	13.4
C.D.	5%	0.57	24.1	2.23
	1%	0.79	—	—

TABLE IV.—Effect of application of fungicide on the control of secondary air borne infection of *Helminthosporium oryzae* and yield of Dharial variety of paddy.

Treatment	Dose	Leaf infection value.	Percentage of infected grain	Yield of grain (in lbs).
Control		33.0	25.4	12.42
Organo-mercurial dust	20 lbs. per acre	9.5	10.3	16.53
Cupravit Blue OB	4 lb per 100 gallons of water	18.8	21.8	12.76
Perenox	4 lbs. per 100 gallons of water	13.6	11.3	15.62
C.D.	5%	0.71	1.06	2.94
	1%	0.99	1.49	—

From the data, it may be concluded that the best and consistent results in reduction of leaf and grain infection and increase of yield have been obtained by dusting with organo-mercuric dusts. Large scale field trials with organo-mercurial dusts are needed to throw further light on the possibility of controlling large scale infection in the field on a practical and economic basis.

SUMMARY

Spraying and dusting of different fungicides namely Perenox, Dithane Z-78 and Perelan for control of secondary infection of *Helminthosporium oryzae* can significantly reduce the intensity of leaf infection. With Perenox best results are obtained when two sprays are applied before flowering.

Spraying of Perenox on paddy grown in large blocks in the cultivators' fields results in reduction of leaf infection and grain and increase in yield of grain.

Of the three fungicides, namely, organo-mercuric dust, Perenox (spray) and Cupravit Blue OB (spray) applied on two varieties of paddy in Coochbehar farm, West Bengal, best and consistent results were obtained by application of organo-mercurial dust which resulted in lowering of leaf and grain infection and increase of yield.

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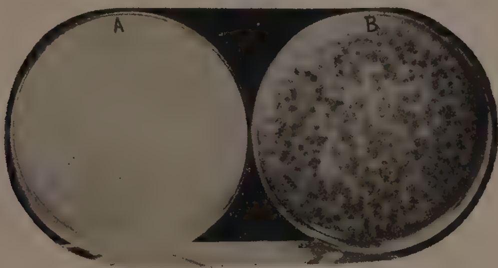
Phytopathological Notes

Bacteriophage specific for Xanthomonas Citri (Hasse) Dowson, the citrus canker organism—M. K. Hingorani, B. C. Dave and Nirmaljit Singh. A bacteriophage producing lysis of *Xanthomonas citri* (Hasse) Dowson, the causal organism of citrus canker, was isolated from the diseased leaves of *Citrus paradisi* (grape fruit), collected from the orchard of the Horticulture Division of the Indian Agricultural Research Institute (Plate I).



A. Control plate showing continuous dense growth of *X. citri*.
B. Plate showing plaques produced by the phage.

The phage had a maximum "Titre" of 10^{-9}, having 7 x 10^9 phage particles per ml. of the stock phage. The selective liquid medium, consisting of $MgS0_4$. $7H_20$ (0.02%), Kcl (0.02%), $(NH)_4H_2P0_4$ (0.1%,) yeast extract (0.04%), and glucose (1.0%), and nutrient agar (having 1 %agar), adjusted to pH 7.0 using M/7.5 phosphate buffer, were found to be the best media for lysis and plaque formation, respectively. The minimum, optimum and maximum temperatures for lysis were found to be 10°, 25–27° and 35°C., respectively while the corresponding temperatures for plaque formation were 10°, 27–30° and 35°C. The optimum pH at which the lytic principle was found to be most active was 7.5 for both lysis and plaque formation. The phage was inactivated by a 10-minute exposure at 75°C. and by chloroform treatment when 0.5 ml. of the chemical was added to 5 ml. of the stock phage. The phage was found to be host-specific as it did not attack any of the other 19 species of the genus *Xanthomonas* tested. Secondary cultures of the test organism almost invariably appeared after some days in the liquid cultures lysed by the action of the lytic principle as also in the phage-treated media plates showing plaque formation.

The writers wish to record their grateful thanks to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his keen interest and helpful suggestions.

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Preliminary note on the occurrence of the Yellow-net vein disease of Mulberry. S. P. Raychaudhuri, S. N. Chatterjee and H. K. Dhar. The indigenous mulberry species viz., *Morus indica* L. and *Morus alba* L. were introduced around 1946 at the Central Sericultural Research Sub-station at Kalimpong. Since then, these are being grown in this farm for feeding the mulberry silkworm (*Bombyx mori* Linnaeus) for rearing work. Recently some foreign varieties of mulberry have also been introduced in this farm as well as in the two newly established sericultural farms viz., Foreign Race Seed Station and Hill Nursery of the Department of Commerce and Industries, Government of West Bengal. During the course of survey carried out periodically in these farms, occurrence of yellow-net vein was observed for the first time in a few *Morus indica* plants raised from cuttings (Fig. 1) in the month of April, 1958, at the Central Sericultural Research Sub-station at an altitude of about 3,500 feet. It was further observed that the disease slowly spreads to mulberry plants grown on different terraces in this farm and it was also noticed in the mulberry variety Kalimpong Selected in May, 1959. Subsequently, the disease was observed in two Japanese mulberry plants, variety Koksu, at the Foreign Race Seed Station at an altitude of 4,200 feet. Later during further survey work the occurrence of the disease was noted in *M. indica* plants at the Matigara Sericultural Nursery, Siliguri (District Darjeeling), West Bengal Sericultural Nursery, Ranaghat (District Nadia), farms of the Central Sericultural Research Station, and West Bengal Sericultural Nursery at Berhampore (District Murshidabad) and in a private farm at Piasbari in Malda District (West Bengal). Very recently symptoms of yellow-net vein resulting in chlorotic areas starting from the margins of affected leaves was observed in *M. indica* at the Central Sericultural Research Sub-station, Kalimpong.

Yellow-net Vein of Mulberry (*Morus indica* L.)

Yellow-net vein resembling the yellow-net vein of mulberry described herein has also been observed on *Hibiscus rosa-sinensis* L. at the Central Sericultural Research Sub-station, State Agricultural Farm and farm of

St. Joseph's School at Kalimpong, on *Croton sparsi-flora* at the Central Sericultural Research Sub-station, Kalimpong and at the Matigara Sericultural Nursery, Siliguri, and on *Poinsettia pulcherrima* Grah. at Kalimpong.

Since the disease appeared to be of virus origin, attempts were made to transmit the disease from *M. indica* to *M. indica* by various methods of grafting. Most of the wedge-grafts, root-grafts and bud-grafts were found to be unsuccessful and failed to transmit the disease while only one out of 18 pen-grafts developed disease symptoms on new growth. Thereafter, attempts were made to transmit the disease by inarch-grafting employing healthy and diseased plants of *M. indica*. It was observed that by this method seven out of 22 successful grafts (nearly 33 per cent of the plants) developed typical symptoms of yellow-net vein disease thereby proving that this is a virus disease.

Attempts are being made to establish the vector of the virus causing the disease and transmission tests with white-flies collected on azalea which occur in this area, aphids on maize and *Hibiscus rosa-sinensis* L. are being carried out. While-flies are not of common occurrence in this region and on very careful survey of the locality, white-flies on azalea have been observed in some nurseries and gardens.

The occurrence of the yellow-net vein disease happens to be the first record of the incidence of virus disease of mulberry in India. So far three virus diseases of mulberry viz.; virosis, dwarf and mosaic diseases (Endo and Kurasawa, 1937; Kawai, 1939; Yakohama, 1958) have been reported.

It may incidentally be mentioned that the problem of mulberry diseases may have a direct bearing on the development of improved races of silk-worms and production of silk in the country. It has been established (Yakohama, 1958) that mulberry leaves affected by powdery mildew (*Phyllactinia corylea* (Pers.) Karst) are less nutritive and apt to dry fast. Such leaves are unsuitable for the purpose of feeding the silk-worm and frequently cause bad cocoon crop in summer and autumn. Since mulberry happens to be the only food plant of the mulberry silkworm the question whether malnutrition of mulberry silkworms is apt to be caused if the latter are fed on mulberry leaves infected with yellow-net vein disease requires consideration.

Thanks are due to Dr. R.S. Vasudeva, Joint Director and Head of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, for his continued interest in the work and helpful criticisms and to the Indian Council of Agricultural Research for financing the scheme.

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Immunity to Papaya Mosaic Virus in the Genus Carica—S. P. Capoor and P. M. Varma. A devastating virus disease of papaya (*Carica papaya* L.) has been prevalent all over the central belt of India extending from East to West and seriously affecting cultivation of papaya (Capoor and Varma, 1958). In some parts of Deccan, where commercial papaya cultivation was a flourishing industry at one time, the disease has been responsible for complete annihilation of large commercial and prosperous papaya plantations.

The papaya mosaic is caused by a virus transmitted mechanically and in nature by *Myzus persicae* Sulz., *Aphis malvae* Koch., *Aphis gossypii* Clover, *Aphis medicaginis* Koch., and also by *Macrosiphum sonchi* L. The first three aphid species are efficient vectors and are mainly responsible for the spread of the virus in plantations. The virus is not seed-borne, but is thermo-labile, unstable and also non-persistent (Capoor and Varma, 1958).

During 1945–47 concerted efforts were made to eradicate the disease from papaya plantations by systematic rogueing of diseased plants within a five mile radius around the Agricultural College Estate in Poona and by a regular inspection of new plantations. Also, as an experimental measure papaya plantation extending over 20 acres in Theur about 14 miles from Poona was sprayed from October to December every week with a mixture of 5 per cent DDT (Dichlorodiphenyle-trichlorethane) and fish-oil rosin soap in addition to systematic rogueing of diseased plants. None of these attempts, however, gave a satisfactory control of the mosaic disease mainly because of recurrence of fresh infections. The failure to control the disease was, therefore, attributed to the following factors:

(i) plantation growers and individual kitchen garden owners were usually reluctant to rogue out diseased papaya plants in good time;

(ii) even isolated diseased papaya plants at considerable distances served as effective foci of infection; and

(iii) the insecticidal spraying was ineffective since the aphid vectors do not colonise on papaya plants in nature. Aphids are only casual visitors to papaya and during their flight and short feeding they transmit the mosaic virus from diseased to healthy papaya plants. Also DDT spraying did not give any relief but only helped the mites to multiply profusely on sprayed plants and, thus, indirectly caused severe mite injury which adversely affected the crop.

It, therefore, appeared that the only possible method of eliminating the ravages of the disease and protecting the papaya crop was to search for source of resistance amongst the cultivated papaya varieties available in India and abroad, and also among the wild *Carica* species. With this object in view papaya varieties Gokeralla, Goda Kawala, Madhu Bindu,

C.P. 124, Ranchi, Kaajimuindeen, Ceylon, Bombay, Poona Long, Poona Round, American, Washington Special Hawaiian, Solo Hawaii, Hortus Gold, Mammoth, Giant, Peterson, Bettina, Philippine, Paradenia, Honey Dew, Solo line-8, Venezuela and Coorg Honey were tested but all were found to be susceptible.

Seeds of *Carica cauliflora*, *C. candamarcensis*, *C. microcarpa* and *C. guoodotana* were obtained from Venezuela and seedlings raised under insect proof condition were tested against the mosaic virus by mechanical juice inoculation. Except. *C. cauliflora* which was not infected, all others were found to be susceptible (Vasudeva, 1959).

Several batches of *C. cauliflora* seedlings were inoculated both through infective juice by mechanical inoculation and through *Myzus persicae* and *Aphis gossypii*, the vectors of papaya mosaic virus (Capoor and Varma, 1958). Each *C. cauliflora* seedling was infested with at least 50 adults of the viruliferous aphids twice but none of the inoculated *C. cauliflora* seedlings was infected. Also none of the inoculated *C. cauliflora* seedlings yielded papaya mosaic virus on back inoculation to healthy seedlings of *C. papaya*.

These observations proved that *Carica cauliflora* is immune to infection by the papaya mosaic virus. Whether this immunity is gene-controlled or brought about as a result of an inhibitor of virus multiplication present in leaves of *C. cauliflora* is not yet known.

Malaguti *et al* (1957) observed that *C. cauliflora* and *C. candamarcensis* were resistant to papaw mosaic virus in Venezuela, but *C. candemercensis* is highly susceptible to the papaya mosaic in India.

The authors, are indebted to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for suggesting the problem and for his guidance during the progress of these experiments and to S. Horovitz, Faculty of Agronomy, Maracay, Venezuela, for supplying the seed of *Carica* spp.

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The Perithecia Producing Mutant of Macrophoma mangiferae.— M. K. Hingorani, T. S. Reddy, Nirmal Jit Singh and H. S. Sohi. During the cultural studies of *Macrophoma mangiferae*, the causal organism of blight disease of mango, and its ultraviolet light induced mutant, it was found that the mutant strain when grown on mango leaf decoction agar 25°–30°C. produced small, round and dark brown perithecia. From these perithecia, a large number of single ascospore cultures were established on oatmeal agar, the growth of which was similar to that of the mutant. Morphologically also they resembled the mutant strain. Since ascospore culture successfully infected mango (varieties *Chowsa*, *Dasheri*, *Langra* and *Sundhuri*), *Eugenia jambolina*, *Eryobotrya japonica*, *Ficus carica* and *Vitis vinifera*, which were also found susceptible to *Macrophoma mangiferae* and its mutant in artificial inoculations. Figure shows infection produced by the three isolates (Parent, mutant and single ascospore culture) on the leaves of the mango variety *Dasheri*. Thus, the connection of the ascigerous stage, obtained in culture, with the conidial stage was proved in all respects.

The perithecia start as loose hyphal wefts, dark brown in colour, which soon become sclerotioid in nature. These are half embedded in the substratum. The perithecia are formed singly and are globose, ostiolate, without any beak or clypeus. These are true representatives of *Sphaeriaceae*. Contents of the perithecium are hyaline to olivaceous within, with abundant protoplasmic contents. These measure 330–735 μ in width and 255–630μ in length. Paraphyses or paraphysoids are absent. Asci are clavate, slightly narrowed above, tapering below, shortly stipitate, octcsporus and measure 70–115 x 75–14μ (Fig. 2). Ascospores are hyaline, single

Fig. 1. Successful reproduction of the disease symptoms on the leaves of mango (var. *Dasheri*) by artificial inoculations with (A) Parent, (B) Mutant & (C) Single ascospore culture.

Fig. 2. Asci and ascospores from a single perithecium (X430).

celled, pear shaped with granular contents, arranged obliquely biseriate within the ascus and are $13.5 - 2.5 \times 6–9\mu$ in size. The fungus is considered to be a species of *Phomatospora*. This identification is, however, tentative and requires further support in the morphological characters on the host as the absence of paraphyses and paraphysoids is yet to be confirmed. The presence of these structures on the natural habitat will ultimately decide its affinites with the genus *Physalospora* which is knwon to be the perfect stage of several species of *Macrophoma*.

The perithecial stage was obtained from a single ascospore culture, thus proving the homothallic nature of the fungus.

The writers are grateful to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Patholcgy, for his keen interest and for correcting the manuscript; to Mr. Ram Lal Munjal, Assistant Prof. of Plant Pathology, for helpful suggestions.

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A method for Screening Wheat Plants for resistance to Neovossia indica—B. L. Chona, R. L. Munjal and K. L. Adlakha. Partial bunt (popularly known as Karnal bunt) caused by *Neovossia indica*(Mitra) Mundkur is widely prevalent in Northern India. It causes substantial loss to the wheat crop in Punjab, Western U.P. and Delhi in certain years. The detailed symptoms of the disease have been described by Mitra (1935) and its air-borne nature has been proved by Mundkur (1943).

The brand spores of the fungus get into the soil either at the time of threshing or as external seed contaminants and germinate towards the middle of February to mid-March, when suitable soil temperature and moisture are available. Each brand spore produces a short or long germ tube to come up to the soil level and bears as many as 110–185 primary sporidia at its tip. These sickle shaped sporidia are wafted in the air by the wind and are lodged on the wheat flowers. The flowering season and sporidial formation synchronize if favourable temperature and moisture for spore germination are present. The sporidia germinate on the wheat flowers and enter the developing grain through the ovary wall. The extent to which the endosperm of the grain is utilized by the fungus and converted into bunt spores depends on the environmental conditions subsequent to infection. Partial destruction of grain is the rule and complete destruction is an exception. The mycelium of the fungus penetrates the emrybonic tissue but does not do any damage. Some of the partially bunted grains have been observed to germinate and produce a normal healthy plant.

In smuts or bunts, where floral infection takes place, Moore's vaccuum method for inoculation has been invariably used. The other methods such as opening a floret by hand with forceps and adding a droplet of

spore suspension with the help of dropper or dusting the floret with smut spores have also been tried. The last method has, however, not given encouraging results.

As stated earlier, the Karnal bunt fungus enters the developing ovary through the ovary wall and not through stigma and style as in the case of loose smut of wheat or barley. It was, therefore, considered desirable to inject spore or sporidial suspension with the help of a hypodermic syringe into the boot-leaf before the ear emerges out of it, in addition to the other methods generally adopted for smuts or bunts with floral mode of infection. The spore suspension was prepared by taking the bunted grains and crushing them in water. The broken grain pieces were removed by straining the mixture through a thin muslin cloth, care being taken to see that there were approximately 20–30 spores per drop. A uniform sporidial suspension was made in distilled water by taking the inoculum from one month old culture of *Neovossia indica*, grown on dextrose plus yeast extract, or cowdung extract.

The relative efficacy of the three methods was tested by inoculating the wheat ears by Moore's method, Dropper method and the Boot-leaf inoculation method. In the first two the inoculations were made at the anthesis stage while in the third method, the inoculations were made in boot-leaf stage and the procedure adopted for inoculating the wheat ears was as follows: The inoculum is filled in the hypodermic syringe which is kept in the right hand with the thumb on the piston handle with the needle end facing downward. The boot-leaf of the wheat plant is supported with left hand and a prick made in the boot-leaf in its upper half. The piston is then pressed slowly and the inoculum goes into the boot-leaf and fills the empty space in between the wheat ear and the leaf whorl. The needle is then withdrawn and the puncture made closes itself automatically owing to the elasticity of the boot-leaf wall tissue.

About 50 ears of each of the four selected wheat varieties were inoculated by all the three methods in February, 1957. The results obtained are given below:—

Wheat variety	Per cent infection on ear basis					
	Moore's method		Dropper method		Boot-leaf method	
	Spore suspension	Sporidial suspension	Spore suspension	Sporidial suspension	Spore suspension	Sporidial suspension
NP 165 ...	4.1	31.4	5.7	45.0	32.5	86.0
NP 720 ...	4.8	22.7	4.6	29.2	24.2	80.0
NP 744 ...	2.8	30.0	1.5	31.5	31.5	71.0
NP 760 ...	2.4	11.1	1.8	18.4	28.6	59.0

Boot-leaf inoculation of wheat plants.

The boot-leaf inoculations proved more successful as compared to the other two methods employed. It might, however, be mentioned that the stage of development of the boot-leaf is an important factor. It was observed that in very young boot stage, inoculations were not so successful because of the difficulty of adequate inoculum getting into the compact boot. This method of inoculation is very easy to operate and not so time consuming. Moreover, it is not necessary to cover the inoculated ears with paper bags to maintain required humidity for infection, as is essential in the other methods.

Grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division for his keen interest, encouragement and valuable guidance.

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A new Seedling blight of guava and its control—I. N. Tandon. During July, 1960 a severe seedling blight disease of guava (*Psidium guajava* L.) was observed in the green house of the Institute, where the guava hybrids were raised for experimental purpose and a large number of them died due to this disease. Isolations from the diseased leaves and stems consistently gave rise to *Rhizoctonia* sp. which has not, hitherto, been reported on this host.

The disease mainly manifests itself on seedlings up to 4 months of age, older ones being not affected. It is rarely observed in dry weather and if it appears, its spread is very slow. It is most severe in July-August when the seedlings have luxuriant leafy growth and the weather is humid. The first symptoms of the disease appear on the leaves as small, circular to irregular, brownish spots which spread very rapidly under humid conditions, covering the entire lamina and the petiole. The upper leaves are the first to show the symptoms and the disease progresses from upward to downward. As the disease advances, the leaf drops and within 2 to 3 days it may involve the leaves, petiole, and stem and finally the whole plant dies (Fig. 1). On the petiole and stem elongated dark brown lesions are found.

Frequently strands of coarse mycelium are seen traversing the leaf surface. Under favourable conditions of humidity, the hyphae spread from leaf to leaf, petiole and stem, occasionally tying a number of young leaves together with a bunch of tenacious mycelial strands. The mycelial growth is mostly inconspicuous in dry weather except on dead leaves on which a whitish cobweb like growth may be seen. The mycelium spreads on the tender parts of the green stem and the petiole which later form numerous black sclerotia. These sclerotia appear as very small white hyphal knots, gradually turning brown and finally black. They are first soft, but become hard later on.

An experiment to control the disease was conducted by spraying the plants with Vitigran (copper oxychloride) – 0.4 and 0.5 per cent, Ziram (zinc dimethyl dithiocarbamate) – 0.2 and 0.3 per cent, Ferbam (Ferric dimethyl dithiocarbamate) – 0.2 and 0.3 per cent, Dithane Z-78 (Zinc ethylene bis dithiocarbamate) – 0.2 and 0.3 per cent and Flit 406 (N-trichloromethyl thio – 4 – cyclohexene – 1, 2 – dicarboximide) – 0.2

and 0.3 per cent. Artificial inoculations with the fungus were carried out on the leaves of treated as well as untreated (control) plants 10 days after spraying, whereas observations regarding the incidence of the disease were taken 16 days after spraying. Flit 406 – 0.2 per cent and Ferbam 0.3 per cent were found to be most effective in controlling the disease.

Further details of the disease including control measures, which are still in progress, will be reported later.

The author wishes to express his sincere thanks to Dr. L. B. Singh, Director, Horticultural Research Institute, Saharanpur for his keen interest and encouragement and to Shri J. N. Seth for pointing out the disease and providing the diseased material.

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A note on Schizaphis graminum (Rondani), an additional vector of sugarcane mosaic in India—M. L. Seth and B. L. Chona. *Rhopalosiphum maidis* (Fitch) = *Aphis maidis* Fitch was for the first time experimentaly established as vector of sugarcane mosaic in India by Chona and Seth (1958). Further attempts were made to find out additional vectors of the virus, as in other sugarcane growing countries more than one species of aphids, besides *Rhopalosiphum maidis*, are known to be responsible for the spread of this disease. Smith (1957) has listed no less than 5 species of aphids to be the vectors of sugarcane mosaic virus.

The 'green bug', *Schizaphis graminum* (Rondani) = *Toxoptera graminum* Rondani, has been found to colonize on certain grasses and other graminaceous crops, like wheat and barley, in Delhi during the spring, was tested whether it could transmit sugarcane mosaic virus. The 'green bug' has also been reported by Ingram and Summers (1938) as a vector of minor importance of sugarcane mosaic virus in the United States.

Schizaphis graminum was colonised on wheat and barley plants in the insect-proof cages, to raise the healthy colonies of the insect. Groups of apterous aphids were taken from these colonies and starved for about 4 hours and then transferred to mosaic-infected sugarcane plants of variety *Surkha Saharanpuri* (*Saccharum officinarum* type) for infection—feeding for about 30 minutes, and were then transferred to healthy test-plants of the same cane variety raised under insect-proof conditions. About 30 aphids were put on each test plant, which were kept in small insect-proof chambers. The insects were allowed to feed on the test-plants for 24 hours, after which the plants were removed from the chambers, sprayed with 0.1 per cent solution of Ekatox to kill the aphids, and placed on the glass-house benches and observed for mosaic infection. The results of the transmission tests are presented in the table given below. It usually took 3-4 weeks for the symptoms to appear in the inoculated plants. All the tests were carried out in the insect-proof glass-house and suitable controls were kept in which no case of mosaic was observed.

Transmission tests with *Schizaphis graminum*

Inoculum	Test plant	No. of plants inoculated	No. infected	Percentage of infection
Mosaic infected sugarcane variety *Surkha Saharanpuri*	Sugarcane variety *Surkha Saharanpuri*	40	10	25.0
—do—	Sugarcane variety Co. 313.	27	5	18.5

From the data presented above it is apparent that *Schizaphis graminum* (Rondani) can act as vector of sugarcane mosaic, and is considered to be an additional vector of the disease in India in addition to *Rhopalosiphum maidis* already established.

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Internal Mould of Chillies caused by Alternaria tenuis Auct.— R. L. Mathur and J.P. Agnihotri. Fruit rot of Chillies (*Capsicum annum* L.) due to internal mould resulting in discolouration of the fruits, thereby reducing the market value of the produce is very common in different parts of Rajasthan. An examination of the lots in the markets has revealed the severity to be from 5 to 85 per cent.

Isolations made from surface sterilized (two minutes dip in 0.1 per cent mercuric chloride solution, followed by washings with sterile water) moulded material yielded invariably the fungus *Alternaria* and a pure culture was obtained by single spore technique. The growth and morpho-

logical characters were studied on four media. The hyphae are septate, branched irregularly, hyaline when young, changing to deep olive grey with age. The conidiophores are straight, erect or irregularly bent, 46.8μ in average length, having catenate conidia at the apex. Conidia are light brown, muriform, having rounded bases and tapering gradually towards apex which may be drawn into a non septate beak. They measure 26.4μ x 8.7μ on an average on potato dextrose agar with or without beaks, the latter varying from 3.6 to 7.2μ when present. These characters coupled with number of cross walls confirm it to be the fungus *Alternaria tenuis* Auct. In pathogenicity tests typical discolouration and growth of the mould was observed only in injured fruits.

Since the fungus is a wound parasite, mechanical injury to the fruits is essential for the infection.

Careful picking of the fruits from the plants, with special attention being given to avoid breaking of the pedicels, transporting the fruits in containers (baskets) with loose packing to avoid hard pressing and breaking of the fruits and storing the produce in comparatively warmer and airy place as the fungus is favoured by lower temperatures (16–26 °C), would probably help in reducing losses due to this mould.

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Bacterial Leafspot diseases of Eleusine coracana and Setaria italica in Madras State,—G. Rangaswami, N. N. Prasad and K. S. S. Eswaran. *Eleusine coracana* Gaertn., *Ragi* or Finger millet and *Setaria italica* Beauv., *Tenai* or Italian millet are two of the important millet crops of India. During September—December 1960 severe leafspot diseases of the two crops cultivated in Chidambaram taluk of South Arcot district, Madras State, were found and the diseases were investigated in detail.

On *ragi* the spots appear on the upper as well as lower surface of the leaf blade. They are linear and spread along the veins. They are 2 to 4 mm. long, but often extend upto an inch or more. In the beginning the spots are light yellowish brown, but soon become dark brown. In advanced stages the leaf splits along the streak, giving a shredded appearance. All the leaves, including the tender shoots, in a plant are affected (Fig. 1). The bacterium appears to mainly affect the leaves, but at times characteristic streaks may be found on the peduncle of the earhead. These streaks are very narrow, 5 to 10 mm. in length and appear subcuticular.

The organism was brought into pure culture and its pathogenicity on *E. coracana* was established in the usual manner. It was also studied for its morphological, cultural and biochemical properties and on the basis

of the results obtained named as a new species. There is no earlier report of any bacterial disease on *ragi* in India or in any other country.

Xanthomonas eleusineae Rangaswami, Prasad and Eswaran, sp. nov.

The bacterium is a short rod, 1.8–2.7 x 0.8–1.0μ, single, monotrichous with a single polar flagellum, aerobic, gram-negative, non-capsulated, non-spore forming and non-acid fast. It forms dull yellow slimy and shiny colonies on nutrient agar and growth in nutrient broth is turbid with pellicle formation. Gelatin is rapidly liquefied but starch is not utilized. Litmus milk turns neutral and gets coagulated. Nitrate is reduced, H_2S produced but no ammonia and indole production. It gives positive lipolytic activity and negative M.R. and V.P. tests. It utilizes lactose as a carbon source, with acid production and little or no gas formation.

The bacterium is pathogenic on *Eleusine coracana* Gaertn., causing minute leafspots and leaf-stripes. On artificial inoculation the bacterium was found to infect *Zea mays* L., but not *Oryza sativa* L., *Setaria italica* Beauv., *Sorghum vulgare* Pers., *Pennisetum typhoideum* Rich., *Panicum maximum* Jacq., *Saccharum officinarum* L. and *Paspalum scrobiculatum* L.

On *tenai* the disease first manifests itself as minute water-soaked spots, which in 2 or 3 days develop into light brown spots, about 1 mm. in width and 1 to 2 mm. in length. The spots are somewhat irregular in shape, with yellowish margins and uneven colouring of the spot. Several spots coalesce to form large patches, which sometimes cover the whole width of the leaf blade, excepting the mid-rib (Fig. 2). Older leaves are more commonly affected, the young leaves on the plant being quite free from the symptoms. With the advancement of the disease the affected leaves may dry in patches but do not drop off. The stem, earhead etc. do not seem to be affected by the organism.

The causal bacterium was brought into pure culture and its pathogenicity on *tenai* was established in the usual manner. In other countries *Pseudomonas setariae* (Okabe) Savulescu and *Ps. alboprecipitans* Rosen are known to infect this host (Elliott, 1951). But when the organism was studied for its morphological, cultural and biochemical properties it was found to be much distinct from the other two reported from outside India and so it was identified as a new bacterium.

Xanthomonas indica Rangaswami, Prasad and Eswaran, sp. nov.

The bacterium is a short rod, 1.8–2.7 x 0.8–1.0μ, single or in chains of two, monotrichous with single polar flagellum, aerobic, gram-negative non-capsulated, non-spore forming and non-acid fast. It forms yellowish brown spreading colonies on nutrient agar and growth in nutrient broth is turbid with profuse yellow sedimentation. No soluble pigment is proudced either in agar or liquid media. Gelatin is moderately liquefied but starch is not hydrolysed. Litmus milk turns highly alkaline but without any coagulation. Nitrate is reduced and ammonia and H_2S produced;

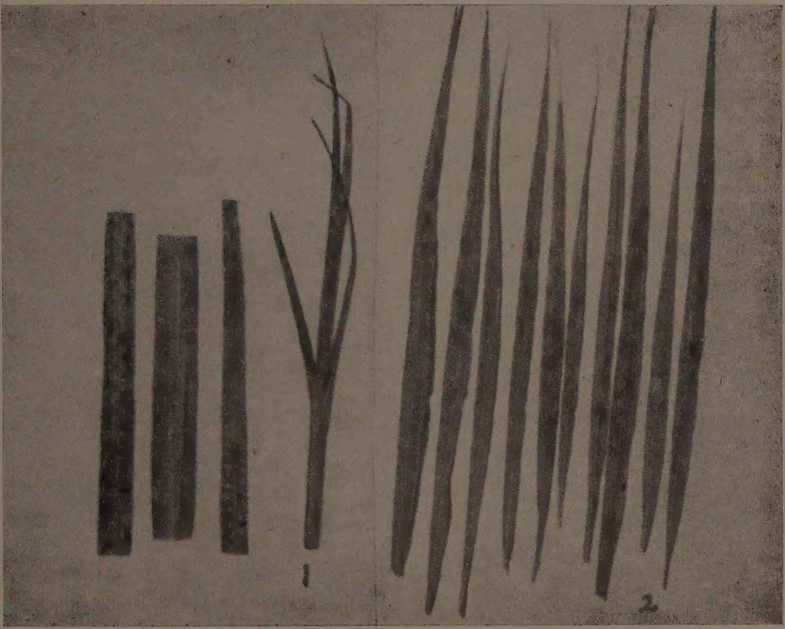

Fig. 1. Leafspot of ragi caused by *X. eleusineae.*

Fig. 2. Leafspot of tenai caused by *X. indica.*

positive lipolytic activity but no indole production; negative M. R. and positive V. P. tests. Lactose is utilized as a carbon source, with slight acid and no gas production.

The bacterium is pathogenic on *Setaria italica* Beauv., causing minute linear spots, which coalesce to form large patches. On artificial inoculation it infects *Zea mays* L., and *Sorghum vulgare* Pers., but not *Brachiaria mutica* Stapf., *Cenchrus ciliaris* L., *C. glauca* L., *C. setigerus* Vahl., *Eleusine coracana* Gaertn., *Oryza sativa* L., *Panicum maximum* Jacq., *P. miliaceum* L., *Paspalum scrobiculatum* L., *Pennisetum typhoideum* Rich., and *Saccharum officinarum* L.

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Contributions should be on one side of the page, double spaced, with a 1¼th inch margin on the left. In form and style, such as punctuation, spelling and use of italics, the manuscript should conform to the best Journals in the U.K. and U.S.A. Authors should strive for a clear and concise style of writing. The name and address of the Institution at which the work was done should be cited immediately after the SUMMARY at the end of the article on left hand side. Tables should be numbered and each table should have a heading stating briefly its contents. References to literature should be made as foot notes *only* when four or fewer citations are given. If there are more, they should be listed under 'REFERENCES' at the end of the paper and referred to by date in brackets in the body of the article. Citation should give the name of the author (or authors), his (or their) initials, year of publication and then the full title correctly, followed by the name of the Journals, number of the volume, a colon and page numbers. If the title is in a foreign language, then diacretic signs and capitalization should be precisely as in the original. The names of the Journal should be as abbreviated in the WORLD LIST OF PERIODICALS, 2nd Ed., 1934, but as the book may not be available to all, contributors are requested to give the titles in full. Abbreviating will in that case, be done by the Editors. If an article has not been seen in original, then that fact should be clearly stated. An example citing is given below:—

Conovor. R.A. (1948)......Studies of two viruses causing mosaic disease of soyabean. *Phytopathology,* 38 : 724-735.

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Authors are invited to consult Bisby's 'An Introduction to Taxonomy and Nomenclature of Fungi' (1945), p.p. 38-41 and Riker's "The preparation of manuscripts for Phytopathology, *Phytopathology* 36 : 953-977, 1946, before preparing their mss. and figures.

Articles will be published in the order of their approval for publication but the address of the retiring President and invitation articles will be published when received.

To comply with the International Rules of Botanical Nomenclature, Latin descriptions must be supplied to validate new species and genera.

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(*Continued from cover 2nd page*)

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